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Report on the 10th inter-laboratory comparison organised by the European Union Reference Laboratory for Polycyclic Aromatic Hydrocarbons

Four marker PAHs in chocolate and cocoa butter

Radoslav Lizak, Zuzana Zelinkova, Philippe Verlinde, and Thomas Wenzl

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Polycyclic Aromatic Hydrocarbons

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Four marker PAHs in chocolate and cocoa butter

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EC-JRC-IRMM
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1 Summary

This report presents the results of the tenth inter-laboratory comparison (ILC) organised by the European Union Reference Laboratory for Polycyclic Aromatic Hydrocarbons (EU-RL PAHs) on the determination of the four EU marker PAHs, benz[*a*]anthracene (BAA), benzo[*a*]pyrene (BAP), benzo[*b*]fluoranthene (BBF) and chrysene (CHR), in cocoa products, particularly in plain chocolate and cocoa butter. It was conducted under ISO Standard 17043 accreditation.
In agreement with National Reference Laboratories, the test material used in this exercise were commercial products. Participants also received a solution of PAHs in solvent of their choice (either toluene or acetonitrile) with disclosed PAH content for the verification of their instrument calibration.
The test materials were value assigned by applying robust statistics to the values reported by the participants and were used to benchmark their results.
Both officially nominated National Reference Laboratories (NRLs) and official food control laboratories (OCLs) of the EU Member States were admitted as participants.
The participants were free to choose the method of analysis. The four EU marker PAHs were chosen as target analytes as limits for their sum were recently introduced in European legislation. The performance of the participating laboratories in the determination of the target PAHs in cocoa butter and plain chocolate was expressed by z-scores. Additionally, the compliance of reported method performance characteristics was checked against specifications given in legislation.

A summary of the performance of the participants in the determination of the four marker PAHs in the two test materials is given in the following table.

Participant group	Reporting laboratories	Calculated z-scores	z-scores ≤ |2|	z-scores ≤ |2|
	#	#	#	%
NRLs	25	362	296	82
OCLs	22	325	260	80

Some laboratories reported results that seem to be systematically biased. It is therefore recommended to investigate this further.

2 Introduction

The Institute for Reference Materials and Measurements (IRMM) of the European Commission's Joint Research Centre operates the European Union Reference Laboratory for Polycyclic Aromatic Hydrocarbons in Food (EU-RL-PAH). One of its core tasks is to organise inter-laboratory comparisons (ILCs) for the National Reference Laboratories (NRLs) [1, 2].

Polycyclic aromatic hydrocarbons (PAHs) constitute a large class of organic substances. The chemical structure of PAHs consists of two or more fused aromatic rings. PAHs may be formed during the incomplete combustion of organic compounds and can be found in the environment. In food, PAHs may be formed during industrial food processing and domestic food preparation, such as smoking, drying, roasting, baking, frying, or grilling.

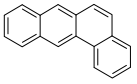
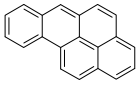
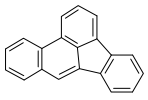
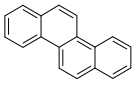
In 2002 the European Commission's Scientific Committee on Food identified 15 individual PAHs as being of major concern for human health. These 15 EU priority PAHs should be monitored in food to enable long-term exposure assessments and to verify the validity of the use of the concentrations of benzo[*a*]pyrene (BAP) as a marker for a "total-PAH content" [3]. The toxicological importance of these compounds was confirmed in October 2005 by the International Agency for Research on Cancer (IARC), which classified BAP as carcinogen to human beings (IARC group 1), cyclopenta[*cd*]pyrene - CPP, dibenzo[*a,h*]anthracene - DHA, and dibenzo[*a,l*]pyrene - DLP as probably carcinogenic to human beings (group 2a), and nine other EU priority PAHs as possibly carcinogenic to human beings (group 2b) [4].

As a consequence, the European Commission (EC) issued Commission Regulation (EC) No 1881/2006 setting maximum levels of benzo[*a*]pyrene in food, Commission Regulation (EC) No 333/2007 laying down sampling methods and performance criteria for methods of analysis for the official control of benzo[*a*]pyrene levels in foodstuffs, and Commission Recommendation 2005/108/EC on the further investigation into the levels of PAHs in certain foods [5, 6, 7].

To evaluate the suitability of BAP as a marker for occurrence and toxicity of PAHs in food, the European Commission asked the European Food Safety Authority (EFSA) for a review of the previous risk assessment on PAHs carried by the Scientific Committee on Food (SCF).

The scientific opinion on PAHs in food was published by EFSA in June 2008 [8]. EFSA concluded that benzo[*a*]pyrene was not a suitable indicator for the occurrence of PAHs in food and that four (PAH4) or eight PAHs (PAH8) were more suitable indicators for the occurrence of PAHs in food. However, PAH8 does not provide much added value compared to PAH4. Following these conclusions the Standing Committee on the Food Chain and Animal Health agreed to base risk management measures on four PAHs (PAH4) - BAA, BAP, BBF, and CHR. However, maximum levels for BAP would be maintained to ensure comparability with historical data. In the following the PAH4 will be also indicated as "the four EU marker PAHs". They are listed in **Table 1**. A maximum level for the sum of the four PAHs was included in the amendment of Commission Regulation (EC) No 1881/2006 [6]. Coherently, also Commission Regulation (EC) No 333/2007 [7] which lays down minimum method performance criteria was revised by Commission Regulation (EC) No 836/2011.

Table 1: Names and structures of the four EU marker PAHs.

1	**Benz[*a*]anthracene (BAA)**		**2**	**Benzo[*a*]pyrene (BAP)**	
3	**Benzo[*b*]fluoranthene (BBF)**		**4**	**Chrysene (CHR)**	

3 Scope

As specified in Regulation (EC) No 882/2004 on official controls performed to ensure the verification of compliance with food and feed law, animal health and animal welfare rules [2], one of the core duties of EU-RLs is organising inter-laboratory comparison tests (ILCs).
This inter-laboratory comparison study aimed to evaluate the comparability of analysis results reported by National Reference Laboratories (NRLs) and EU official food control laboratories (OCLs) for the four EU marker PAHs in cocoa products. The appropriateness of the reported measurement uncertainty was also tested as this parameter is important in the compliance assessment of food with EU maximum levels.
The ILC was designed and evaluated under the umbrella of IRMM's accreditation according to ISO Standard 17043:2010 [9].

4 Participating Laboratories

Officially nominated NRLs and OCLs of the EU Member States were admitted as participants. The participants are listed in **Table 2** and **Table 3** respectively.

Table 2: List of participating National Reference Laboratories

Institute	***Country***
AGES - Österreichische Agentur für Gesundheit und Ernährungssicherheit, Kompetenzzentrum Cluster Chemie	AUSTRIA
Scientific Institute of Public Health	BELGIUM
SGL - State General Laboratory, Environmental and other Food Contamination Laboratory	CYPRUS
Nàrodní referenční laboratoř pro polycyklické aromatické uhlovodíky - Státní veterinární ústav Praha	CZECH REPUBLIC
Division of Food Chemistry, National Food Institute, Technical University of Denmark	DENMARK
Veterinary and Food Administration, Chemical Laboratory	DENMARK
Tartu Laboratory of Health Protection Inspectorate	ESTONIA
EVIRA - Finnish Food Safety Authority	FINLAND
LABERCA - Laboratoire d'Etude des Résidus et des Contaminants dans les Aliments	FRANCE
BVL - Bundesamt für Verbraucherschutz und Lebensmittelsicherheit	GERMANY
GCSL - General Chemical State Laboratory - Food Division - Laboratory	GREECE
Central Agricultural Office, Food & Feed Safety Directorate, Food Residues Toxicological Dept.	HUNGARY
Central Agricultural Office, Food and Feed Safety Directorate, Feed Investigation NRL	HUNGARY
The Public Analyst's Laboratory Dublin	IRELAND
Istituto Superiore di Sanità	ITALY
BIOR - Institute of Food Safety, Animal Health and Environment	LATVIA
National Veterinary Laboratory (National Food and Veterinary Risk Assessment Institute)	LITHUANIA
RIKILT- Institute of Food Safety	NETHERLANDS
NIFES - National Institute of Nutrition and Seafood Research	NORWAY
National Institute of Public Health - National Institute of Hygiene	POLAND
IP – INRB -Instituto Nacional dos Recursos Biológicos	PORTUGAL
SVUPUDK - State Veterinary and Food Institute Dolný Kubín	SLOVAKIA
Zavod za zdravstveno varstvo Maribor	SLOVENIA
AESAN - Centro Nacional de Alimentaciòn (Spanish Food Safety and Nutrition Agency)	SPAIN
SLV - Livsmedelsverket	SWEDEN
FERA - The Food and Environment Research Agency	UNITED KINGDOM

One of the 26 NRLs did not at all report results for this PT, whereas a second NRL reported only a very limited number of results. Both NRLs reasoned this with instrument break downs.

Table 3: List of participating Official Food Control Laboratories

Institute	*Country*
Institut für Umwelt und Lebensmittelsicherheit, Bregenz	AUSTRIA
Analytec - Labor für Lebensmitteluntersuchung und Umweltanalytik ZT-GmbH	AUSTRIA
LARECO	BELGIUM
Vlaamse Instelling voor Technologisch Onderzoek VITO	BELGIUM
Federal Laboratory for the Safety of the Food Chain	BELGIUM
LDA56	FRANCE
LVD72	FRANCE
LDA22	FRANCE
IDAC	FRANCE
SCL Massy	FRANCE
LEAV	FRANCE
Eurofins WEJ Contaminants GmbH	GERMANY
Landesuntersuchungsamt Bremen	GERMANY
CVUA Münsterland-Emscher-Lippe	GERMANY
Landesbetrieb Hessisches Landeslabor	GERMANY
CVUA Stuttgart	GERMANY
Landeslabor Berlin-Brandenburg	GERMANY
Landesamt für Verbraucherschutz Sachsen-Anhalt	GERMANY
Institut für Hygiene und Umwelt, Hamburg	GERMANY
Thüringer Landesamt für Lebensmittelsicherheit und Verbraucherschutz	GERMANY
CVUA Ostwestfalen-Lippe	GERMANY
Minton Treharne and Davis Ltd	UNITED KINGDOM
Staffordshire Scientific Services	UNITED KINGDOM
Worcestershire Scientific Services	UNITED KINGDOM
City of Edinburgh Council	UNITED KINGDOM

Twenty two of the 25 registered OCLs reported results.

5 Time frame

The ILC was agreed with the NRLs at the EU-RL PAH workshop in Brussels on the 6th of April 2012. It was announced on the IRMM web page (see ANNEX 1) and invitation letters were sent to the laboratories on the 29th of May 2012 (see ANNEX 2). Test samples were dispatched (see ANNEX 3) on the 16th of July 2012 and the deadline for reporting of results was set to the 14th of September 2012. However, two NRLs were not able to report before 30th of September.
The documents sent to the participants are presented in ANNEX 4.

6 Confidentiality

The identities of participants are kept confidential unless the participant provides a letter of consent to the PT organiser giving permission to disclose his/her details and results to a third party.

7 Test materials

7.1 Preparation

The test materials of this PT round were cocoa butter (CB) and plain dark chocolate (CHOC). This matrices are representative for the food category 6.1.2 "Cocoa beans and derived products" specified in Commission Regulation (EC) No 836/2011, with a maximum level for BAP and for the sum of the four PAHs (in the following indicated as SUM4PAH) of 5.0 µg/kg and 35.0 µg/kg respectively. Both maximum levels will come into force on 1 April 2013.

Participants also received a solution of the 15+1 EU Priority PAHs in either acetonitrile or toluene (according to their choice, see ANNEX 3) with disclosed concentrations, which allowed them to check their instrument calibration against an independent reference. Participants received the technical specifications (see ANNEX 5) of the chosen solution together with the test material.

The cocoa butter test material was prepared at the EU-RL PAH laboratories from four kilos of commercial cocoa butter, which was supplied by a Belgian Chocolatier. The whole quantity of cocoa butter was melted at 60 °C in a water bath, until the liquid became completely clear, and further stirred over night, before aliquots of about 20 mL were bottled into amber glass crimp cap vials. The chocolate test material was acquired at local supermarkets. In total 5 kg of plain dark chocolate of a particular brand with labelled cocoa and fat contents of minimum 86 % and 56.5 % respectively were purchased and cryo-grinded. The fine particles were further homogenized by stirring and by repetitive application of a sample divider. Aliquots of about 30 g of the chocolate test sample were packed into amber glass screw cap vials.
The standard solution was prepared from neat certified reference materials (purchased from BCR®, Institute for Reference Materials and Measurements, Geel, Belgium, except CPP - purchased from Biochemisches Institut für Umweltkarzinogene, Großhansdorf, Germany, BCL - purchased from Dr. Ehrenstorfer, Germany, and DIP - purchased from Campro Scientific, Germany). Single standard stock solutions of each analyte were produced by substitution weighing of neat substance on a microbalance and dissolution in toluene. Mixed standards were prepared gravimetrically from the single standard stock solutions in the respective solvents and further diluted to the concentrations specified in ANNEX 5. The standard solutions were ampouled under inert atmosphere and flame sealed in 5 ml amber glass ampoules.

7.2 Homogeneity and stability

Homogeneity of the test samples was evaluated according to ISO Standard 13528 [10]. An external laboratory was contracted for the analysis of the cocoa butter test material. The homogeneity study of the chocolate test material was performed in-house.
Ten units of each of the test materials were selected randomly and analysed by gas chromatography hyphenated to mass spectrometry. The test materials were rated sufficiently homogeneous (see ANNEX 6).

The stability of the test material was evaluated by analysing the test material after the deadline for reporting of results. Significant differences of the analyte contents between the analysis results before dispatch and after expiration of the reporting deadline were not found. Hence stability of the samples over the whole study period can be assumed.

7.3 Assigned value and standard deviation for proficiency assessment

The robust estimates of the mean values of the results of the participants, according to ISO 13528 Algorithm A, were applied as assigned values for the proficiency assessment [11]. Results that were reported as below a certain value were excluded from the calculations. The H15 estimate is gained by iteration, where in each iteration step pseudo mean values and pseudo variances of the data are calculated. In the following iteration step extreme values (outside the range pseudo mean ± a certain multitude of the pseudo standard deviation) are replaced by the mean value ± a certain multiple of the standard deviation of the previous step, and new pseudo mean and pseudo variance values are calculated. Via this approach pseudo means and pseudo variances converge rapidly to robust estimates of the mean value and of the variance.
The assigned values of the target PAHs are listed in **Table 4** to **Table 6**. **Table 7** provides the robust mean for the fat determination.
The uncertainties of the assigned values were derived from 1.25 times the robust standard deviation of the reported results, divided by the square root of the number of results included in the calculations [10]. The uncertainty of the SUM4PAH parameter was calculated from the uncertainties of the individual analytes, applying the law of error propagation.
The uncertainties of the contents of the standard solutions covered uncertainties stemming from the purity of the reference materials used and the weighing operation carried-out.

The standard deviation for proficiency assessment, σ_P, was set for the individual analytes equal to the maximum tolerable uncertainty (Uf), which is calculated according to Equation 1 [9]. A LOD value of 0.30 µg/kg, and α equal to 0.2 were applied for this purpose. The standard deviation for proficiency testing was calculated for the SUM4PAH parameter from the σ_P - values of the individual analytes applying the law of error propagation.

Equation 1 $$U_f = \sqrt{(\mathrm{LOD}/2)^2 + (\alpha \mathrm{C})^2}$$

where U_f relates to the maximum tolerated standard measurement uncertainty, LOD to the limit of detection, α to a numeric factor depending on the concentration C as given in Commission Regulation (EC) No 333/2007, amended by Regulation (EC) 836/2011.

Table 4: Analyte contents of the cocoa butter test material

		Assigned value	u	σ_P	
Analyte	**Analyte short name**	**µg/kg**	**µg/kg**	**µg/kg**	**%**
Benz[*a*]anthracene	BAA	1.55	0.07	0.34	22.2
Benzo[*a*]pyrene	BAP	0.86	0.03	0.23	26.6
Benzo[*b*]fluoranthene	BBF	1.20	0.06	0.28	23.6
Chrysene	CHR	2.28	0.15	0.48	21.1
Sum of the four marker PAHs	SUM4PAH	5.89	0.18	0.69	11.8

σ_p standard deviation for proficiency assessment.
u standard uncertainty of the assigned value (k=1).

Table 5: Analyte contents of the chocolate test material, expressed on product basis

		Assigned value	u	σ_P	
Analyte	**Analyte short name**	**µg/kg**	**µg/kg**	**µg/kg**	**%**
Benz[*a*]anthracene	BAA	1.72	0.09	0.38	21.8
Benzo[*a*]pyrene	BAP	0.78	0.04	0.22	27.7
Benzo[*b*]fluoranthene	BBF	1.02	0.06	0.25	24.8
Chrysene	CHR	2.12	0.11	0.45	21.2
Sum of the four marker PAHs	SUM4PAH	5.64	0.16	0.67	11.9

σ_p standard deviation for proficiency assessment.
u standard uncertainty of the assigned value (k=1).

Table 6: Analyte contents of the chocolate test material, expressed on fat basis

		Assigned value	u	σ_P	
Analyte	**Analyte short name**	**µg/kg**	**µg/kg**	**µg/kg**	**%**
Benz[*a*]anthracene	BAA-FAT	2.91	0.17	0.60	20.7
Benzo[*a*]pyrene	BAP-FAT	1.37	0.07	0.31	22.8
Benzo[*b*]fluoranthene	BBF-FAT	1.69	0.10	0.37	21.9
Chrysene	CHR-FAT	3.60	0.20	0.74	20.4
Sum of the four marker PAHs	SUM-FAT	9.57	0.29	0.67	11.9

σ_p standard deviation for proficiency assessment.
u standard uncertainty of the assigned value (k=1).

Table 7: Fat content of the chocolate test material

		Robust mean
Analyte	**Analyte short name**	**%**
Fat content	FAT	54.9

8 Design of the proficiency test

The design of the PT foresaw triplicate analysis of the test samples and reporting on product basis of the individual results of replicate analyses for the single analytes, in the following indicated with the extension REP. Additionally a "value for proficiency assessment", in the following denoted as "final value", was requested, expressed on product basis, for both the single analytes and the sum of the four PAHs. All results had to be reported corrected for recovery (and recovery had to be stated in the questionnaire together with other parameters of the method applied); final results had also to be accompanied by the respective expanded measurement uncertainty and the coverage factor. Additionally, the final results for the chocolate samples had to be reported expressed on fat basis, as required by legislation, and the fat content of the chocolate sample had to be stated.
Only final results were used for performance assessment.
Participants were asked to report besides analysis results also details of the applied analysis method (see ANNEX 7).
Each participant received at least one ampoule of a solution of the target PAHs in the chosen solvent (2 ml), with disclosed content, and at least one crimp cap vial containing the cocoa butter test sample as well as two screw cap vials with the chocolate test material.

9 Evaluation of Laboratories

9.1 General

The most important evaluation parameter was the performance of the laboratories in the determination of the target PAHs in the test materials, which was expressed by z-scores.
The results reported by participants are listed in ANNEX 8. Some results were reported as smaller than a certain threshold value. However all threshold values were significantly lower than the assigned values, indicating that the respective analyses were negatively biased. Therefore, these results were rated assuming the threshold value as content value.

The compliance with legislation of method performance characteristics for the determination of the four marker PAHs was evaluated as well.

9.2 Evaluation criteria

z-Scores

z-Scores were calculated based on the final values. Equation 2 presents the formula for calculation of z-scores.

Equation 2

$$z = \frac{\left(x_{lab} - X_{assigned}\right)}{\sigma_P}$$

where z refers to the z-score, x_{lab} to the reported "value for proficiency assessment", $X_{assigned}$ to the assigned value, and σ_P to the standard deviation for proficiency testing.

The performance of the laboratories was classified according to ISO/IEC 17043:2010 [9]. Following scheme is applied for the interpretation of z-scores:

|score| ≤ 2.0 = satisfactory performance
2.0<|score| < 3.0 = questionable performance
|score| ≥ 3.0 = unsatisfactory performance

9.3 Evaluation of results

z-Scores were attributed only to "values for proficiency assessment" (final values). The individual results of replicate analyses were not rated. Also the results of the fat determination were not rated. However they are presented and discussed further down.

Each laboratory had to report a total of 40 results; therefore the expected number of results of the 48 reporting participants was 1920. They submitted in total 1818 results, which equals to about 95 %.
About 81 % of the results reported by the participants obtained a satisfactory z-score. The percentage of satisfactory scores was slightly higher among the NRLs if compared to OCLs. Lowest number of satisfactory z-scores was attributed to results expressed on fat basis for the chocolate test material, where the bias of the fat determination contributes to the gross bias.

Figure 1 and **Figure 2** provide overviews of the z-scores assigned to the results for the cocoa butter test material for NRLs and OCLs respectively. The z-scores awarded to the results for the chocolate test material are presented in **Figure 3** and **Figure 4** for results expressed on product basis (µg/kg chocolate), and in **Figure 5** and **Figure 6** for results expressed on fat basis (µg/kg fat). The larger the triangles, the larger were the differences to the assigned values. Red triangles indicate z-scores above an absolute value of three, whereas yellow triangles represent z-scores in the questionable performance range. For questionable and unsatisfactory scores, the corresponding score values are presented next to the triangles. Some laboratories had major problems with the determination of the target PAHs in both the cocoa butter and the chocolate test material, e.g. participant 1008 overestimated the BAP contents of the two samples by far. Hence this participant shall scrutinize her/his analysis procedure for severe bias, which might be caused by interferences, or instrument calibration. Participant 6022 did not report a single analysis result within the satisfactory performance range.
The numerical values of the calculated z-scores are compiled in **Table 8** to **Table 13** for both NRLs and OCLs . z-Scores with an absolute value of above 2 are given in bold, red font.

The establishment of proper measurement uncertainty values still caused problems for the SUM4PAH parameter. Fifteen out of 35 participants reported for the sum of the contents, expressed on fat basis, of the four PAHs in the chocolate test material measurement uncertainty values that differed significantly from the values, which were derived by the law of error propagation.
Hence the EU-RL PAHs will continue to pay special attention to this parameter in the ILCs to come, as it has major implications on the assessment of compliance of food with European legislation.

Figure 7 shows the distribution of the reported fat contents. The fat content value corresponding to the centre of the distribution is about 1.7 % below the value given on the packaging. Numerical values of the reported fat contents are given in ANNEX 8.

The graphical representations of the distribution of results for the individual analytes are given in ANNEX 8 together with the results of replicate analyses.
For each analyte the figure shows the individual analysis results of the three replicate determinations. The assigned value is shown as green dotted line. The blue boxes represent the expanded uncertainties (k=2) reported by participants for the "value for proficiency assessment". The arithmetic mean of the results of the individual participant is indicated in the blue boxes by a blue line. The red lines represent deviations from the assigned value of ± 2$\sigma_{p.}$

Figure 1: Graphical presentation of **z-scores** corresponding to the "values for proficiency assessment" reported by the **NRLs** for the contents of BAA, BAP, BBF, CHR, and the SUM4PAH parameter in the ***cocoa butter*** test material.

Blue triangles indicate satisfactory performance; yellow triangles indicate questionable performance; red triangles indicate non-satisfactory performance; z-score values are presented next to the triangles for the last two performance categories.

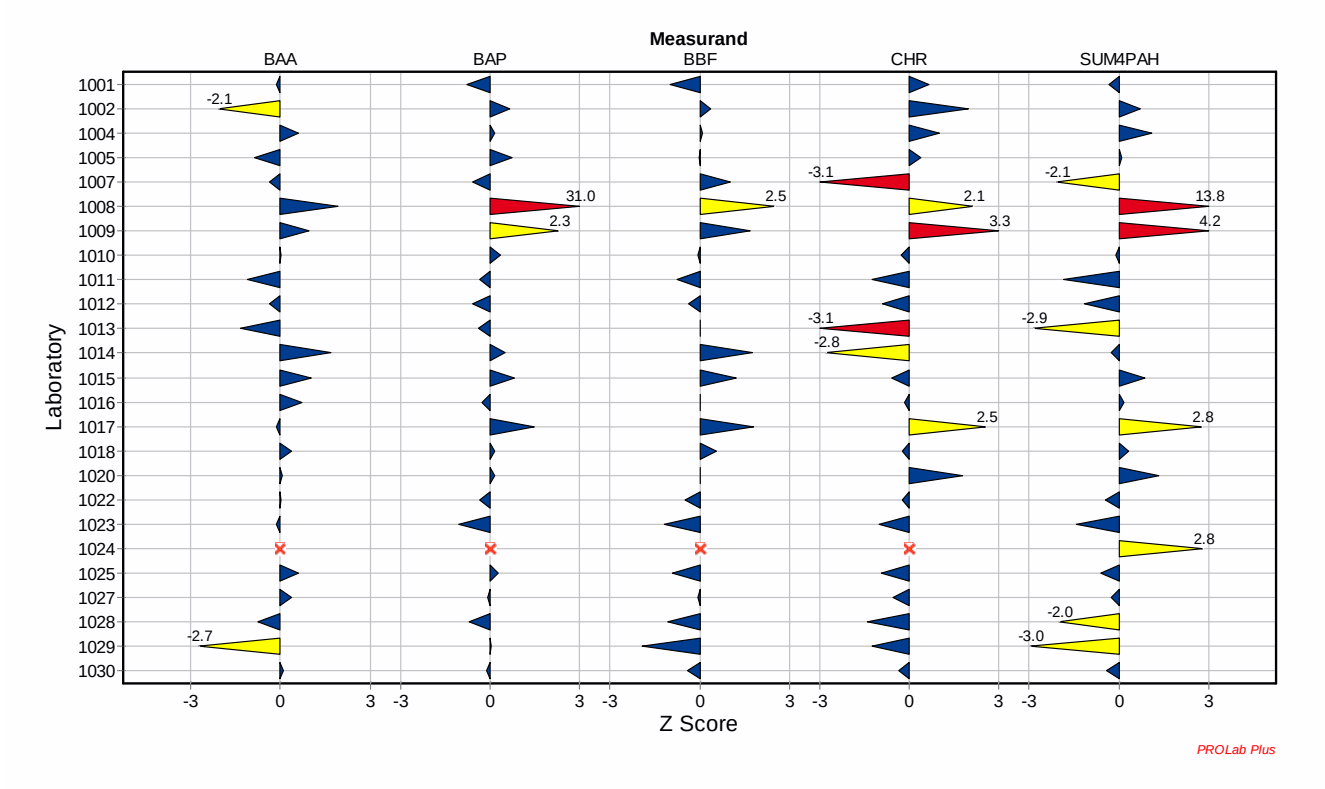


Table 8: Compilation of **z-scores** calculated from the "results for proficiency assessment" reported by the **NRLs** for test material ***cocoa butter*****:** z-scores outside the satisfactory range (|z| > 2) are indicated by bold red font.

	BAA		BAP		BBF		CHR		SUM 4 PAH	
	µg/kg		µg/kg		µg/kg		µg/kg		µg/kg	
Assigned value	1.55		0.86		1.20		2.28		5.89	
σ_P	0.34		0.23		0.28		0.48		0.69	
Participant	Result	z-score	Result	z-score	Result	z-score	Result	z-score	Result	z-score
	µg/kg		µg/kg		µg/kg		µg/kg		µg/kg	
1001	1.5	-0.1	0.68	-0.8	0.92	-1.0	2.6	0.7	5.67	-0.3
1002	0.843	**-2.1**	1.012	0.7	1.303	0.4	3.23	2.0	6.388	0.7
1004	1.76	0.6	0.9	0.2	1.23	0.1	2.76	1.0	6.65	1.1
1005	1.26	-0.9	1.03	0.7	1.19	0.0	2.47	0.4	5.95	0.1
1007	1.42	-0.4	0.73	-0.6	1.49	1.0	0.81	**-3.1**	4.46	**-2.1**
1008	2.2	1.9	8.0	**31.0**	1.9	**2.5**	3.3	**2.1**	15.4	**13.8**
1009	1.88	1.0	1.39	**2.3**	1.67	1.7	3.84	**3.3**	8.78	**4.2**
1010	1.56	0.0	0.94	0.3	1.18	-0.1	2.14	-0.3	5.82	-0.1
1011	1.18	-1.1	0.78	-0.3	0.99	-0.8	1.69	-1.2	4.63	-1.8
1012	1.43	-0.4	0.73	-0.6	1.1	-0.4	1.85	-0.9	5.1	-1.1
1013	1.1	-1.3	0.77	-0.4	1.2	0.0	0.77	**-3.1**	3.91	**-2.9**
1014	2.12	1.7	0.98	0.5	1.69	1.8	0.93	**-2.8**	5.72	-0.2
1015	1.9	1.0	1.05	0.8	1.54	1.2	2.0	-0.6	6.49	0.9
1016	1.8	0.7	0.8	-0.3	1.2	0.0	2.2	-0.2	6.0	0.2
1017	1.5	-0.1	1.2	1.5	1.7	1.8	3.5	**2.5**	7.8	**2.8**
1018	1.68	0.4	0.9	0.2	1.36	0.6	2.17	-0.2	6.11	0.3
1020	1.57	0.1	0.9	0.2	1.2	0.0	3.13	1.8	6.8	1.3
1022	1.557	0.0	0.783	-0.3	1.064	-0.5	2.169	-0.2	5.573	-0.5
1023	1.5	-0.1	0.62	-1.0	0.87	-1.2	1.8	-1.0	4.9	-1.4
1024									7.84	**2.8**
1025	1.75	0.6	0.92	0.3	0.95	-0.9	1.84	-0.9	5.47	-0.6
1027	1.68	0.4	0.84	-0.1	1.18	-0.1	2.01	-0.6	5.71	-0.3
1028	1.3	-0.7	0.7	-0.7	0.9	-1.1	1.6	-1.4	4.5	-2.0
1029	0.62	**-2.7**	0.87	0.0	0.66	-1.9	1.69	-1.2	3.84	**-3.0**
1030	1.58	0.1	0.83	-0.1	1.09	-0.4	2.12	-0.3	5.62	-0.4

Figure 2: Graphical presentation of **z-scores** corresponding to the "values for proficiency assessment" reported by the **OCLs** for the contents of BAA, BAP, BBF, CHR, and the SUM4PAH parameter in the ***cocoa butter*** test material.

Blue triangles indicate satisfactory performance; yellow triangles indicate questionable performance; red triangles indicate non-satisfactory performance; z-score values are presented next to the triangles for the latter performance category.

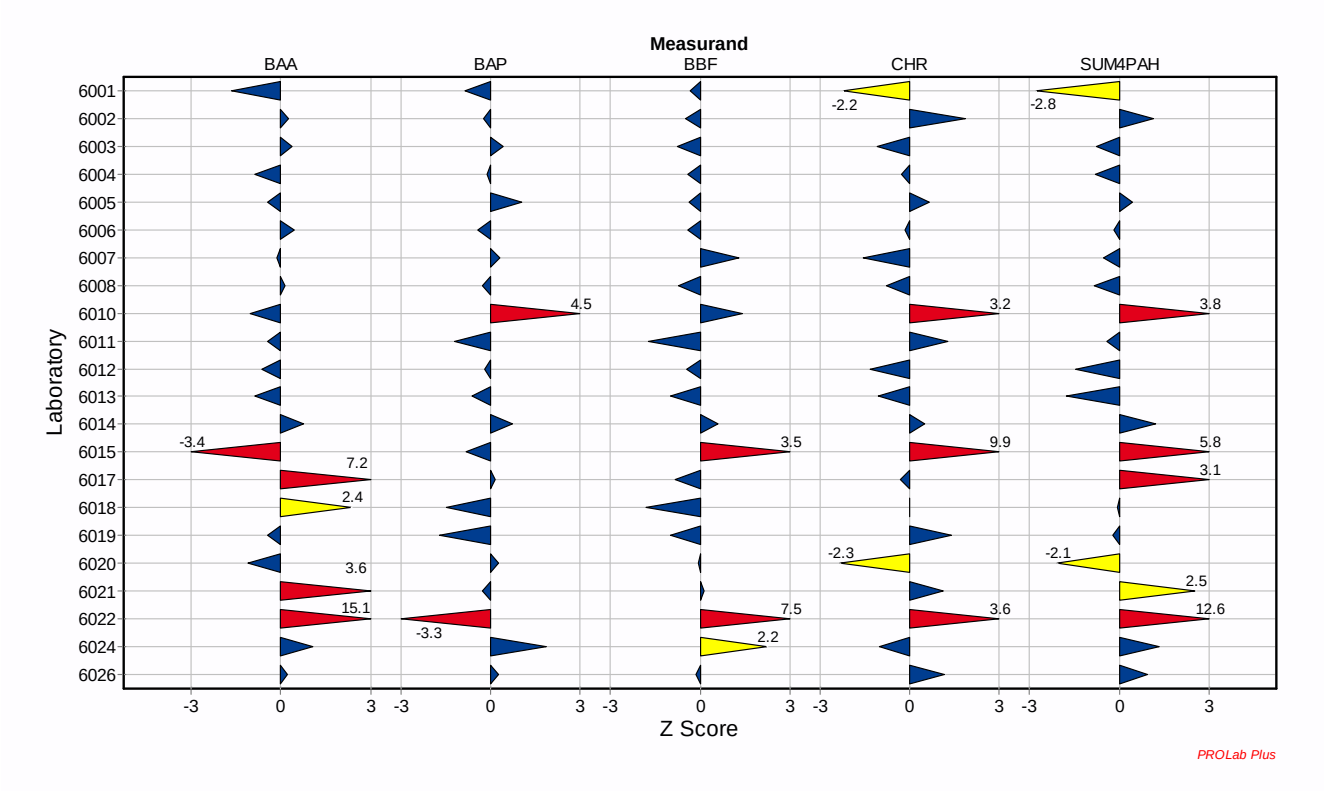


Table 9: Compilation of **z-scores** calculated from the "results for proficiency assessment" reported by the **OCLs** for test material ***cocoa butter***: z-scores outside the satisfactory range (|z| > 2) are indicated by bold red font.

	BAA		BAP		BBF		CHR		SUM 4 PAH	
	µg/kg		µg/kg		µg/kg		µg/kg		µg/kg	
Assigned value	1.55		0.86		1.20		2.28		5.89	
σ_p	0.34		0.23		0.28		0.48		0.69	
Participant	Result	z-score	Result	z-score	Result	z-score	Result	z-score	Result	z-score
	µg/kg		µg/kg		µg/kg		µg/kg		µg/kg	
6001	0.99	-1.6	0.66	-0.9	1.11	-0.3	1.22	**-2.2**	3.98	**-2.8**
6002	1.64	0.3	0.81	-0.2	1.06	-0.5	3.17	1.9	6.68	1.1
6003	1.67	0.4	0.96	0.4	0.99	-0.8	1.75	-1.1	5.37	-0.8
6004	1.26	-0.9	0.83	-0.1	1.09	-0.4	2.15	-0.3	5.33	-0.8
6005	1.4	-0.4	1.1	1.0	1.1	-0.4	2.6	0.7	6.2	0.4
6006	1.706	0.5	0.765	-0.4	1.085	-0.4	2.2	-0.2	5.757	-0.2
6007	1.5	-0.1	0.93	0.3	1.56	1.3	1.54	-1.5	5.53	-0.5
6008	1.6	0.1	0.8	-0.3	1.0	-0.7	1.9	-0.8	5.3	-0.9
6010	1.2	-1.0	1.9	**4.5**	1.6	1.4	3.8	**3.2**	8.5	**3.8**
6011	1.4	-0.4	0.58	-1.2	0.72	-1.7	2.9	1.3	5.6	-0.4
6012	1.34	-0.6	0.82	-0.2	1.08	-0.4	1.65	-1.3	4.89	-1.4
6013	1.26	-0.9	0.722	-0.6	0.923	-1.0	1.78	-1.0	4.68	-1.8
6014	1.81	0.8	1.03	0.7	1.37	0.6	2.53	0.5	6.74	1.2
6015	0.4	**-3.4**	0.67	-0.8	2.19	**3.5**	7.02	**9.9**	9.88	**5.8**
6017	4.01	**7.2**	0.9	0.2	0.97	-0.8	2.13	-0.3	8.01	**3.1**
6018	2.35	**2.4**	0.52	-1.5	0.7	-1.8	2.27	0.0	5.84	-0.1
6019	1.4	-0.4	0.47	-1.7	0.92	-1.0	2.94	1.4	5.73	-0.2
6020	1.18	-1.1	0.92	0.3	1.18	-0.1	1.17	**-2.3**	4.45	**-2.1**
6021	2.78	**3.6**	0.8	-0.3	1.24	0.1	2.81	1.1	7.63	**2.5**
6022	6.7	**15.1**	<0.1	**-3.3**	3.3	**7.5**	4.0	**3.6**	14.6	**12.6**
6024	1.91	1.1	1.29	1.9	1.82	**2.2**	1.8	-1.0	6.82	1.3
6026	1.63	0.2	0.92	0.3	1.16	-0.1	2.84	1.2	6.55	1.0

Figure 3: Graphical presentation of z-scores corresponding to the "values for proficiency assessment" reported by the **NRLs** for the contents of BAA, BAP, BBF, CHR, and the SUM4PAH parameter in the ***chocolate*** test material.
The assessment was based on results expressed on product basis (µg/kg chocolate).
Blue triangles indicate satisfactory performance; yellow triangles indicate questionable performance; red triangles indicate non-satisfactory performance; z-score values are presented above the triangles for the last two performance categories.

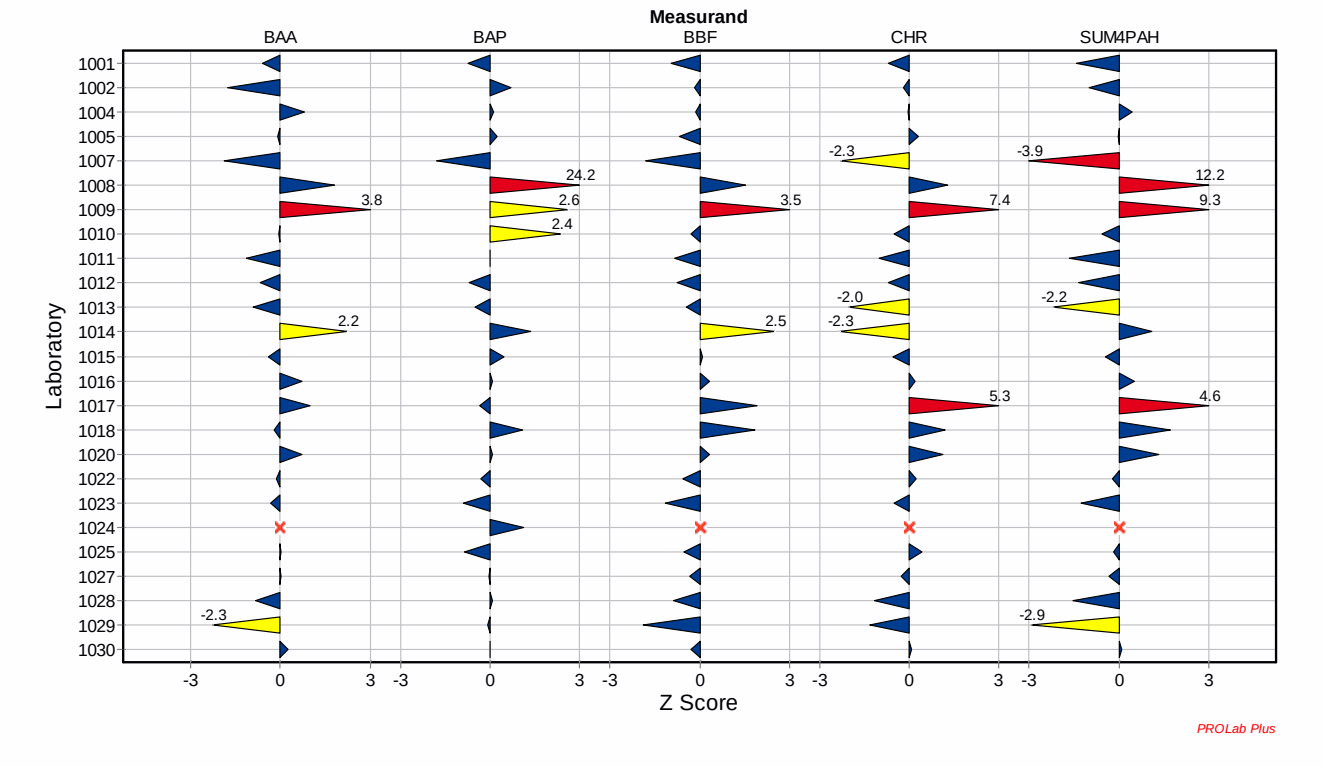


Table 10: Compilation of **z-scores** calculated from the "results for proficiency assessment" reported by the **NRLs** for the ***chocolate*** test material. Results are expressed on product basis (µg/kg chocolate): z-scores outside the satisfactory range (|z| > 2) are indicated by bold red font.

	BAA		BAP		BBF		CHR		SUM4PAH	
	µg/kg		µg/kg		µg/kg		µg/kg		µg/kg	
Assigned value	1.72		0.78		1.02		2.12		5.64	
σ_P	0.38		0.22		0.25		0.45		0.67	
Participant	Result	z-score	Result	z-score	Result	z-score	Result	z-score	Result	z-score
	µg/kg		µg/kg		µg/kg		µg/kg		µg/kg	
1001	1.5	-0.6	0.62	-0.7	0.78	-1.0	1.80	-0.7	4.68	-1.4
1002	1.047	-1.8	0.933	0.7	0.977	-0.2	2.024	-0.2	4.980	-0.9
1004	2.02	0.8	0.81	0.1	0.99	-0.1	2.11	0.0	5.93	0.5
1005	1.68	-0.1	0.83	0.2	0.85	-0.7	2.26	0.3	5.62	0.0
1007	1.00	-1.9	0.39	-1.8	0.57	-1.8	1.09	**-2.3**	3.05	**-3.8**
1008	2.4	1.8	6.1	**24.2**	1.4	1.5	2.7	1.3	13.8	**12.2**
1009	3.17	**3.8**	1.35	**2.6**	1.89	**3.5**	5.46	**7.4**	11.86	**9.3**
1010	1.70	-0.1	1.30	**2.4**	0.95	-0.3	1.89	-0.5	5.26	-0.5
1011	1.29	-1.1	0.78	0.0	0.81	-0.8	1.66	-1.0	4.54	-1.6
1012	1.46	-0.7	0.63	-0.7	0.83	-0.8	1.81	-0.7	4.73	-1.3
1013	1.38	-0.9	0.67	-0.5	0.91	-0.4	1.21	-2.0	4.17	**-2.1**
1014	2.57	**2.2**	1.08	1.4	1.65	**2.5**	1.08	**-2.3**	6.39	1.2
1015	1.56	-0.4	0.88	0.5	1.04	0.1	1.87	-0.6	5.35	-0.4
1016	2.0	0.7	0.8	0.1	1.1	0.3	2.2	0.2	6.0	0.6
1017	2.1	1.0	0.7	-0.4	1.5	1.9	4.5	**5.3**	8.7	**4.6**
1018	1.64	-0.2	1.02	1.1	1.48	1.8	2.66	1.2	6.80	1.8
1020	2.00	0.7	0.80	0.1	1.10	0.3	2.63	1.1	6.53	1.4
1022	1.665	-0.1	0.711	-0.3	0.884	-0.5	2.228	0.2	5.488	-0.2
1023	1.6	-0.3	0.58	-0.9	0.73	-1.2	1.9	-0.5	4.8	-1.2
1024	n.r.		1.03	1.1	n.r.		n.r.		n.r.	
1025	1.73	0.0	0.59	-0.9	0.89	-0.5	2.31	0.4	5.53	-0.1
1027	1.72	0.0	0.77	0.0	0.94	-0.3	1.99	-0.3	5.42	-0.3
1028	1.4	-0.8	0.8	0.1	0.8	-0.9	1.6	-1.2	4.6	-1.5
1029	0.86	**-2.3**	0.76	-0.1	0.55	-1.9	1.52	-1.3	3.68	**-2.9**
1030	1.82	0.3	0.78	0.0	0.95	-0.3	2.15	0.1	5.70	0.1

Figure 4: Graphical presentation of z-scores corresponding to the "values for proficiency assessment" reported by the **OCLs** for the contents of BAA, BAP, BBF, CHR, and the SUM4PAH parameter in the ***chocolate*** test material.
The assessment was based on results expressed **on product basis** (µg/kg chocolate).
Blue triangles indicate satisfactory performance; yellow triangles indicate questionable performance; red triangles indicate non-satisfactory performance; z-score values are presented above the triangles for the latter performance category.

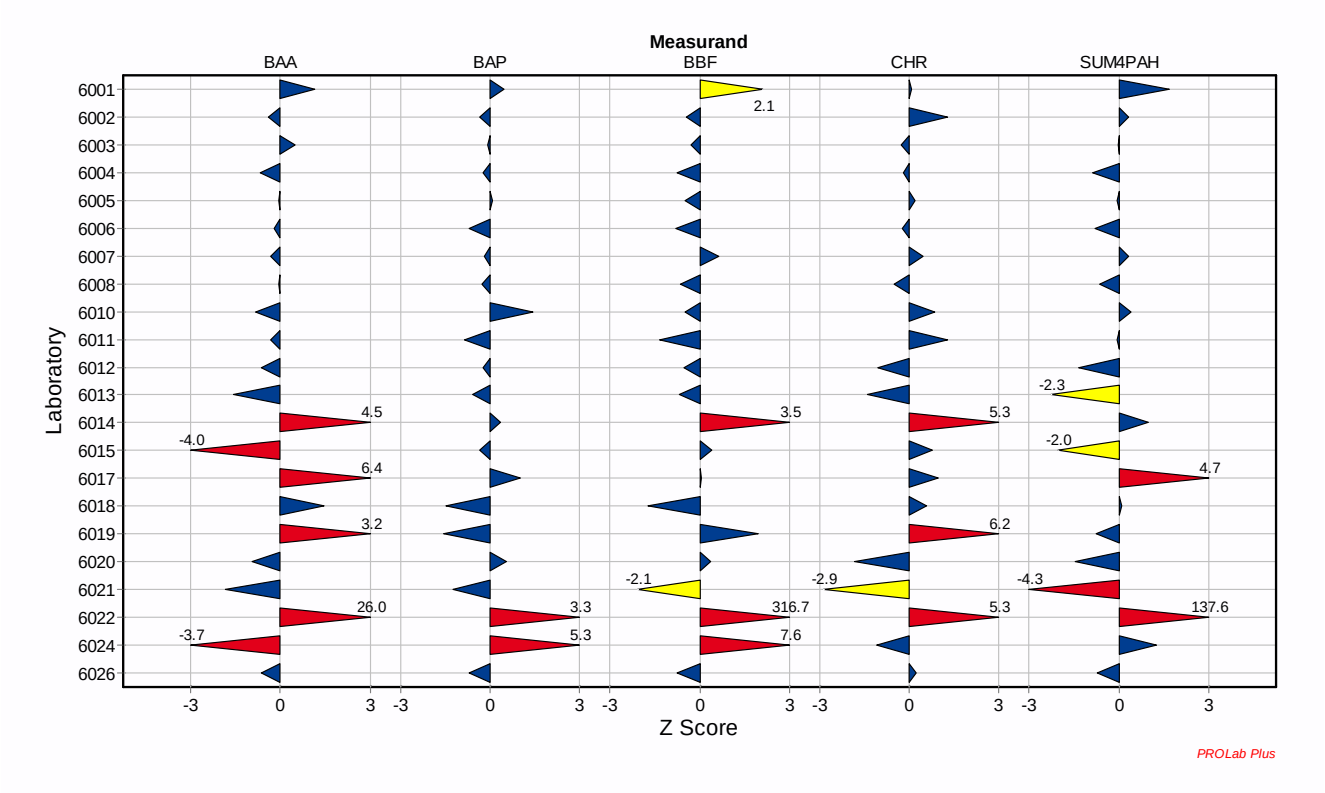


Table 11: Compilation of **z-scores** calculated from the "results for proficiency assessment" reported by the **OCLs** for the ***chocolate*** test material. Results are expressed on product basis (µg/kg chocolate): z-scores outside the satisfactory range (|z| > 2) are indicated by bold red font.

	BAA		BAP		BBF		CHR		SUM4PAH	
	µg/kg		µg/kg		µg/kg		µg/kg		µg/kg	
Assigned value	1.72		0.78		1.02		2.12		5.64	
σ_P	0.38		0.22		0.25		0.45		0.67	
Participant	Result	z-score	Result	z-score	Result	z-score	Result	z-score	Result	z-score
	µg/kg		µg/kg		µg/kg		µg/kg		µg/kg	
6001	2.16	1.2	0.88	0.5	1.55	**2.1**	2.16	0.1	6.76	1.7
6002	1.56	-0.4	0.7	-0.4	0.91	-0.4	2.7	1.3	5.87	0.3
6003	1.91	0.5	0.76	-0.1	0.95	-0.3	2.00	-0.3	5.62	0.0
6004	1.47	-0.7	0.73	-0.2	0.83	-0.8	2.03	-0.2	5.06	-0.9
6005	1.7	-0.1	0.8	0.1	0.9	-0.5	2.2	0.2	5.6	-0.1
6006	1.644	-0.2	0.629	-0.7	0.817	-0.8	2.007	-0.3	5.096	-0.8
6007	1.6	-0.3	0.74	-0.2	1.18	0.6	2.33	0.5	5.85	0.3
6008	1.7	-0.1	0.72	-0.3	0.86	-0.6	1.9	-0.5	5.2	-0.7
6010	1.4	-0.8	1.1	1.5	0.9	-0.5	2.5	0.8	5.9	0.4
6011	1.6	-0.3	0.59	-0.9	0.69	-1.3	2.7	1.3	5.6	-0.1
6012	1.48	-0.6	0.73	-0.2	0.89	-0.5	1.65	-1.0	4.75	-1.3
6013	1.13	-1.6	0.653	-0.6	0.85	-0.7	1.49	-1.4	4.11	**-2.3**
6014	3.44	**4.5**	0.86	0.4	1.9	**3.5**	4.51	**5.3**	6.3	1.0
6015	<0.2	**-4.0**	0.7	-0.4	1.12	0.4	2.46	0.8	4.27	**-2.0**
6017	4.17	**6.4**	1	1.0	1.03	0.0	2.56	1.0	8.8	**4.7**
6018	2.28	1.5	0.46	-1.5	0.59	-1.7	2.38	0.6	5.71	0.1
6019	2.95	**3.2**	0.44	-1.5	1.51	2.0	4.9	**6.2**	5.14	-0.7
6020	1.36	-0.9	0.9	0.5	1.11	0.4	1.3	-1.8	4.67	-1.4
6021	1.02	-1.8	0.51	-1.2	<0.5	**-2.1**	0.82	**-2.9**	2.73	**-4.3**
6022	11.6	**26.0**	1.5	**3.3**	80.2	**316.7**	4.5	**5.3**	97.8	**137.6**
6024	<0.3	**-3.7**	1.94	**5.3**	2.91	**7.6**	1.63	-1.1	6.48	1.3
6026	1.48	-0.6	0.63	-0.7	0.83	-0.8	2.22	0.2	5.16	-0.7

Figure 5: Graphical presentation of z-scores corresponding to the "values for proficiency assessment" reported by the **NRLs** for the contents of BAA_FAT, BAP_FAT, BBF_FAT, CHR_FAT, and the sum of 4 PAHs parameter (SUM_FAT) in the ***chocolate*** test material.
The assessment was based on results expressed on fat basis (µg/kg fat).
Blue triangles indicate satisfactory performance; yellow triangles indicate questionable performance; red triangles indicate non-satisfactory performance; z-score values are presented above the triangles for the last two performance categories.

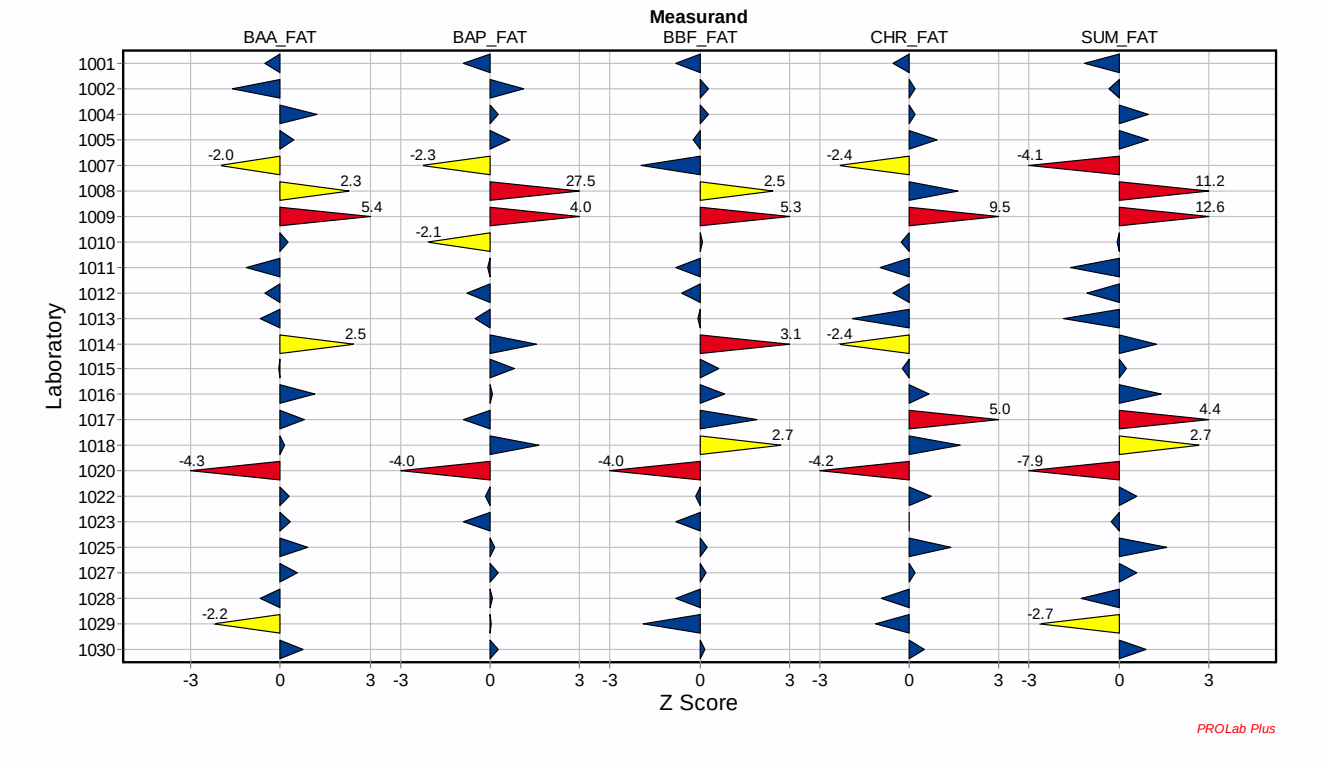


Table 12: Compilation of **z-scores** calculated from the "results for proficiency assessment" reported by the **NRLs** for the ***chocolate*** test material. Results are expressed on fat basis (µg/kg fat): z-scores outside the satisfactory range (|z| > 2) are indicated by bold red font.

	BAA		BAP		BBF		CHR		SUM 4 PAH	
	µg/kg		µg/kg		µg/kg		µg/kg		µg/kg	
Assigned value	2.91		1.37		1.69		3.60		9.57	
σ_P	0.60		0.31		0.37		0.74		1.06	
Participant	Result	z-score	Result	z-score	Result	z-score	Result	z-score	Result	z-score
	µg/kg		µg/kg		µg/kg		µg/kg		µg/kg	
1001	2.6	-0.5	1.1	-0.9	1.4	-0.8	3.2	-0.5	8.34	-1.1
1002	1.937	-1.6	1.720	1.1	1.803	0.3	3.751	0.2	9.210	-0.3
1004	3.64	1.2	1.46	0.3	1.80	0.3	3.73	0.2	10.63	1.0
1005	3.18	0.5	1.57	0.6	1.61	-0.2	4.28	0.9	10.63	1.0
1007	1.70	-2.0	0.66	**-2.3**	0.96	-2.0	1.85	**-2.4**	5.17	**-4.1**
1008	4.3	**2.3**	9.9	**27.5**	2.6	**2.5**	4.8	1.6	21.6	**11.4**
1009	6.15	**5.4**	2.61	**4.0**	3.66	**5.3**	10.61	**9.5**	23.04	**12.8**
1010	3.07	0.3	0.72	**-2.1**	1.72	0.1	3.41	-0.3	9.5	0.0
1011	2.23	-1.1	1.35	-0.1	1.40	-0.8	2.88	-1.0	7.85	-1.6
1012	2.60	-0.5	1.13	-0.8	1.47	-0.6	3.21	-0.5	8.41	-1.0
1013	2.51	-0.7	1.22	-0.5	1.67	-0.1	2.20	-1.9	7.6	-1.8
1014	4.39	**2.5**	1.85	1.5	2.83	**3.1**	1.85	**-2.4**	10.90	1.3
1015	2.88	-0.1	1.62	0.8	1.92	0.6	3.44	-0.2	9.85	0.3
1016	3.6	1.2	1.4	0.1	2.0	0.8	4.1	0.7	11.1	1.5
1017	3.4	0.8	1.1	-0.9	2.4	1.9	7.3	**5.0**	14.3	**4.5**
1018	3.0	0.2	1.87	1.6	2.7	**2.7**	4.86	1.7	12.43	**2.7**
1020	0.36	**-4.3**	0.14	**-4.0**	0.2	**-4.0**	0.47	**-4.2**	1.17	**-7.9**
1022	3.093	0.3	1.320	-0.2	1.642	-0.1	4.136	0.7	10.192	0.6
1023	3.1	0.3	1.1	-0.9	1.4	-0.8	3.6	0.0	9.3	-0.2
1024	n.r.		n.r.		n.r.		n.r.		n.r.	
1025	3.47	0.9	1.42	0.2	1.78	0.2	4.63	1.4	11.3	1.7
1027	3.25	0.6	1.45	0.3	1.77	0.2	3.75	0.2	10.22	0.7
1028	2.5	-0.7	1.4	0.1	1.4	-0.8	2.9	-0.9	8.2	-1.2
1029	1.57	**-2.2**	1.38	0.0	1.00	-1.9	2.78	-1.1	6.72	**-2.6**
1030	3.37	0.8	1.45	0.3	1.75	0.2	3.98	0.5	10.55	1.0

Figure 6: Graphical presentation of **z-scores** corresponding to the "values for proficiency assessment" reported by the **OCLs** for the contents of BAA_FAT, BAP_FAT, BBF_FAT, CHR_FAT, and the sum of 4 PAHs parameter (SUM_FAT) in the ***chocolate*** test material.
The assessment was based on results expressed on fat basis (µg/kg fat).
Blue triangles indicate satisfactory performance; yellow triangles indicate questionable performance; red triangles indicate non-satisfactory performance; z-score values are presented above the triangles for the latter performance category.

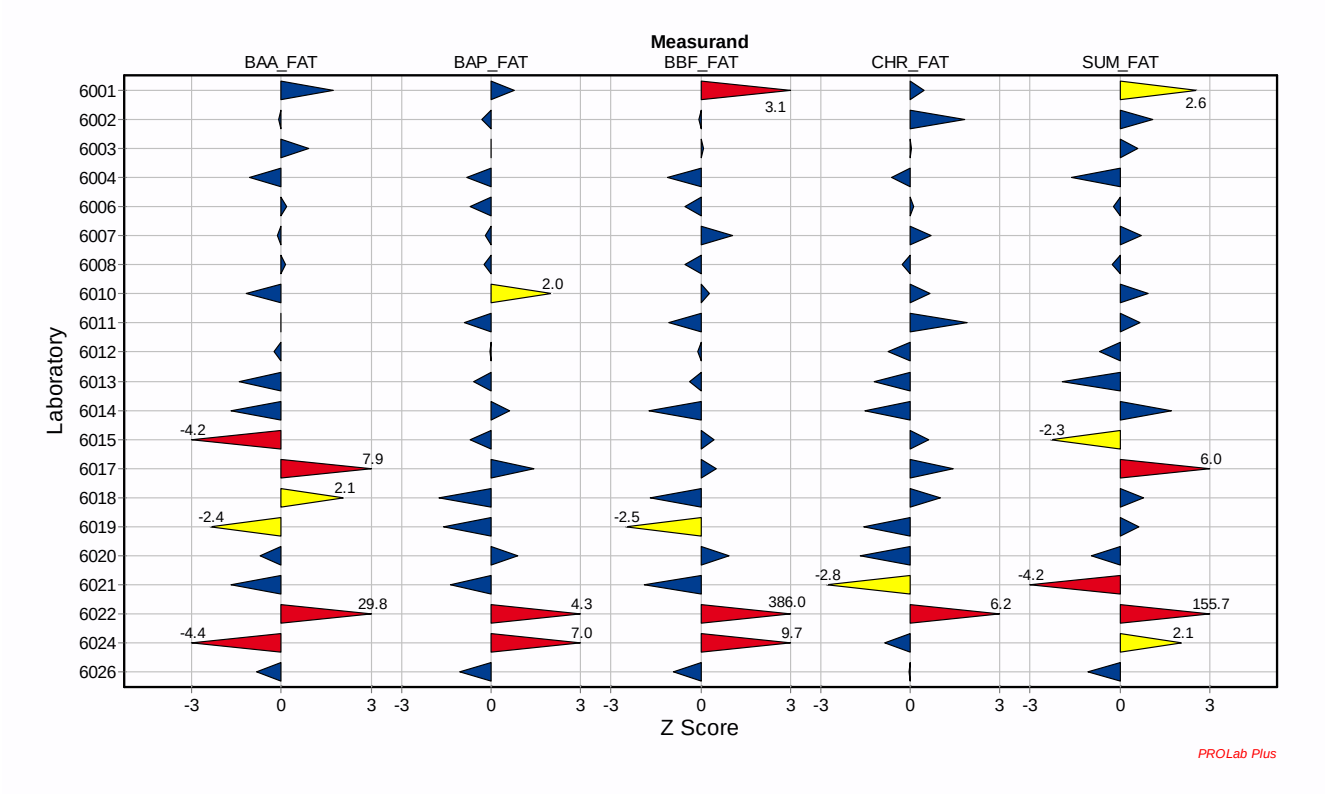
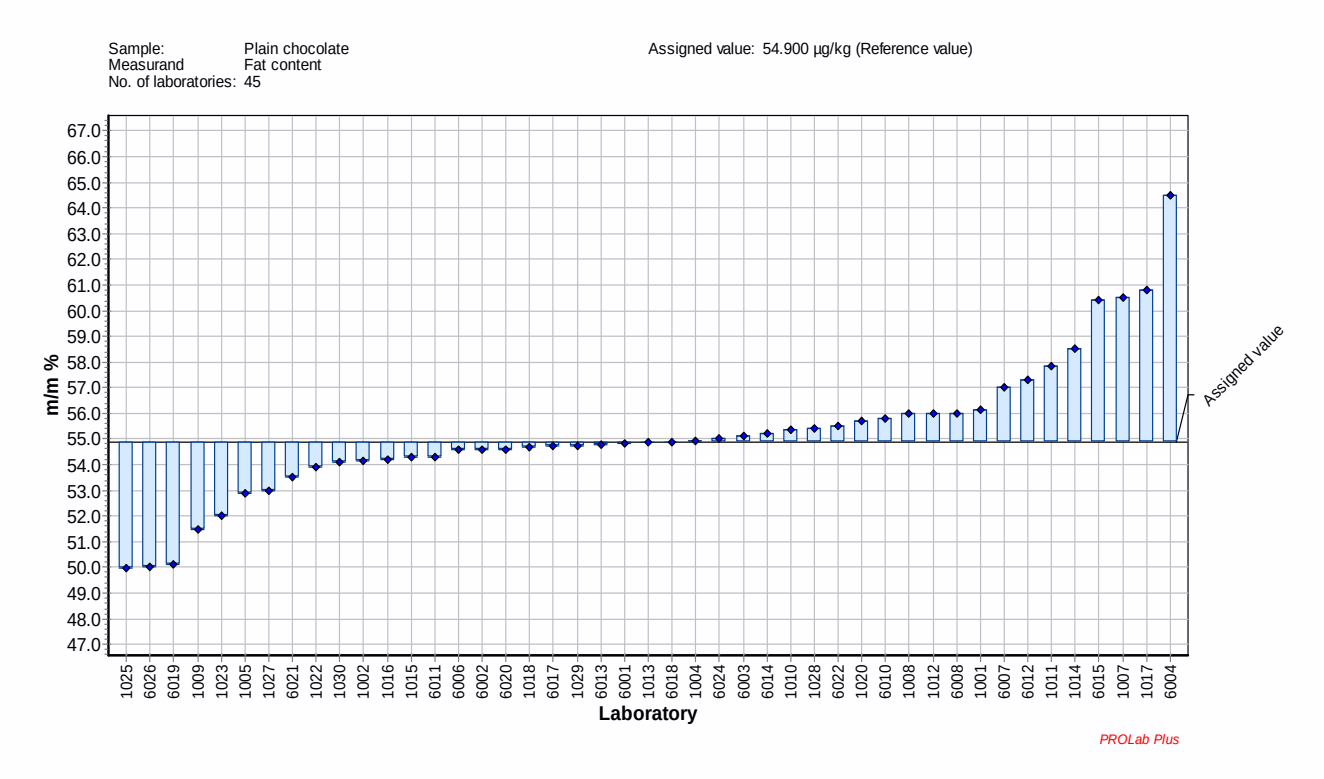


Table 13: Compilation of **z-scores** calculated from the "results for proficiency assessment" reported by the **OCLs** for the ***chocolate*** test material. Results are expressed on fat basis (µg/kg fat): z-scores outside the satisfactory range (|z| > 2) are indicated by bold red font.

	BAA		BAP		BBF		CHR		SUM 4 PAH	
	µg/kg		µg/kg		µg/kg		µg/kg		µg/kg	
Assigned value	2.91		1.37		1.69		3.60		9.57	
σ_P	0.60		0.31		0.37		0.74		1.06	
Participant	Result	z-score	Result	z-score	Result	z-score	Result	z-score	Result	z-score
	µg/kg		µg/kg		µg/kg		µg/kg		µg/kg	
6001	3.95	1.7	1.61	0.8	2.82	**3.1**	3.93	0.4	12.33	**2.7**
6002	2.86	-0.1	1.28	-0.3	1.67	-0.1	4.95	1.8	10.75	1.2
6003	3.47	0.9	1.37	0.0	1.72	0.1	3.63	0.0	10.19	0.6
6004	2.28	-1.1	1.12	-0.8	1.28	-1.1	3.15	-0.6	7.83	-1.6
6005	n.r.		n.r.		n.r.		n.r.		n.r.	
6006	3.01	0.2	1.15	-0.7	1.5	-0.5	3.68	0.1	9.34	-0.2
6007	2.82	-0.2	1.31	-0.2	2.08	1.1	4.12	0.7	10.33	0.8
6008	3.0	0.2	1.3	-0.2	1.5	-0.5	3.4	-0.3	9.3	-0.2
6010	2.2	-1.2	2.0	2.0	1.8	0.3	4.1	0.7	10.6	1.0
6011	2.9	0.0	1.1	-0.9	1.3	-1.1	5.0	1.9	10.3	0.7
6012	2.76	-0.3	1.36	0.0	1.66	-0.1	3.06	-0.7	8.84	-0.6
6013	2.06	-1.4	1.19	-0.6	1.55	-0.4	2.71	-1.2	7.5	-1.9
6014	1.90	-1.7	1.56	0.6	1.05	-1.7	2.49	-1.5	11.4	1.8
6015	<0.4	**-4.2**	1.16	-0.7	1.85	0.4	4.07	0.6	7.08	**-2.3**
6017	7.62	**7.9**	1.82	1.5	1.89	0.5	4.67	1.4	16	**6.1**
6018	4.15	**2.1**	0.83	-1.7	1.07	-1.7	4.34	1.0	10.4	0.8
6019	1.48	**-2.4**	0.88	-1.6	0.76	**-2.5**	2.46	-1.5	10.25	0.7
6020	2.49	-0.7	1.65	0.9	2.04	0.9	2.37	-1.7	8.55	-0.9
6021	1.91	-1.7	0.95	-1.4	1.00	-1.9	1.54	**-2.8**	5.10	**-4.2**
6022	20.8	**29.8**	2.7	**4.3**	144.5	**386.0**	8.2	**6.2**	176.2	**157.2**
6024	<0.3	**-4.4**	3.53	**7.0**	5.29	**9.7**	2.96	-0.9	11.78	**2.1**
6026	2.42	-0.8	1.05	-1.0	1.36	-0.9	3.58	0.0	8.41	-1.0

Figure 7: Graphical presentation of distribution **fat contents** reported for the ***chocolate*** test material.



9.4 Evaluation of compliance with legislation

The characteristics of the methods applied by participants and the results reported are listed in ANNEX 7 and ANNEX 8 respectively.
Compliance with legislation was evaluated on basis of requirements set in Regulation (EC) No 333/2007 as amended by Regulation (EU) No 836/2011 [7]. Non compliant values for LOD, LOQ, and recovery are indicated by bold red font.

In general the number of non-compliances was similar for OCLs and for NRLs. The major shortcoming concerned LODs and LOQs that were higher than the tolerated 0.3 µg/kg and 0.9 µg/kg, respectively. The values for recovery complied with the limits specified in Commission Regulation (EU) No 836/2011. However, it cannot be evaluated whether recovery was understood as yield, as requested, and not as apparent recovery, which might be indicated by recovery values close to 100 %.
Consequently all participants reporting method performance characteristics that do not comply with the minimum performance characteristics specified in legislation shall identify and implement for their analysis methods possibilities for improvement, or shall apply a different, more appropriate analysis procedure.

10 Follow-up actions for underperforming laboratories

All laboratories that got "questionable" or "non-satisfactory" performance ratings are urged to perform root cause analysis, and to implement corrective actions.
The NRLs not reporting were already contacted for explanation.
The EU-RL will set up follow-up measures in due time for all NRLs that received for at least one of the four PAHs (BAA, BAP, BBF, and CHR) z-scores > |3| as required by Regulation (EC) 882/2004, and by the "Protocol for management of underperformance in comparative testing and/or lack of collaboration of National Reference Laboratories (NRLs) with European Union reference laboratories (EU-RLs) activities". These laboratories shall perform as an immediate action root-cause-analysis, and shall report to the EU-RL PAH in writing the identified cause for their underperformance and corrective actions they are going to take. Additionally, they shall participate to an independent (non-EU-RL) proficiency test on the determination of PAHs in food and shall communicate the outcome of this exercise to the EU-RL PAH.

11 Conclusions

Forty-seven participants reported analysis results. The performance of most participants was good. In total 96 % and 88% of the results reported by NRLs and OCLs respectively obtained a satisfactory z-score. However, significant bias can be concluded from the pattern of performance indicators of some laboratories.
A few laboratories did not report measurement uncertainties. They are urged to improve in this respect as this parameter is essential for compliance assessment.
The great majority of participants in this inter-laboratory comparison applied analytical methods which, with regard to performance characteristics, were compliant with EU legislation; however, some participants are encouraged to verify the compliance to legislation of both the method and the modality of data reporting in use at their laboratory.

12 Acknowledgements

The organisers would like to thank Beatriz de la Calle and Franz Ulberth (all from IRMM, Geel, Belgium) for their accurate revision of this report and all NRLs and OCLs for their cooperation.

13 References

1 EU, *COMMISSION REGULATION (EC) No 776/2006 of 23 May 2006 amending Annex VII to Regulation (EC) No 882/2004 of the European Parliament and of the Council as regards Community reference laboratories.* Official Journal of the European Union, 2006. **L 136**: p. 3-8. Available from: http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2006:136:0003:0008:EN:PDF

2 EU, *Regulation (EC) No 882/2004 of the European Parliament and of the Council of 29 April 2004 on official controls performed to ensure the verification of compliance with feed and food law, animal health and animal welfare rules.* Official Journal of the European Communities, 2004. **L191**: p. 1-52. Available from: http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2004:191:0001:0052:EN:PDF

3 *EU, Opinion of the Scientific Committee on Food on the risks to human health of Polycyclic Aromatic Hydrocarbons in food*. (2002). Available from: http://europa.eu.int/comm/food/fs/sc/scf/out153_en.pdf

4 *IARC. Overall Evaluations of Carcinogenicity to Humans.* IARC Monographs on the Evaluation of Carcinogenic Risks to humans (2006). Available from: http://monographs.iarc.fr/ENG/Classification/crthgr01.php

5 *EU, Commission Recommendation (2005/108/EC) of 4 February 2005 on the further investigation into the levels of polycyclic aromatic hydrocarbons in certain foods.* Official Journal of the European Union, 2005. L 34: p. 43-45. Available from: http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2005:034:0043:0045:EN:PDF

6 *EU, Commission Regulation (EC) No 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs.* Official Journal of the European Union, 2006. **L 364**: p. 5-24. Available from: http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2006:364:0005:0024:EN:PDF
Amended by Regulation (EU) No 835/2011 which shall apply from 1 September 2012. Official Journal of the European Union, 2011. **L 215**: p. 4-8

7 *EU, Commission Regulation (EC) No 333/2007 of 28 March 2007 laying down the methods of sampling and analysis for the official control of the levels of lead, cadmium, mercury, inorganic tin, 3-MCPD and benzo(a)pyrene in foodstuffs.* Official Journal of the European Union, 2007. **L 88**: p. 29-38. Available from: http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2007:088:0029:0038:EN:PDF
Amended by Regulation (EU) No 836/2011 which shall apply from 1 September 2012. Official Journal of the European Union, 2011. **L 215**: p. 9-16

8 *EFSA, Scientific Opinion of the Panel on Contaminants in the Food Chain on a request from the European Commission on Polycyclic Aromatic Hydrocarbons in Food*. The EFSA Journal, 2008. 724: p. 1-114. Available from: http://www.efsa.europa.eu/EFSA/efsa_locale-1178620753812_1211902034842.htm

9 *ISO/IEC 17043:2010 (2010)*. Conformity assessment -- General requirements for proficiency testing providers. Geneva, Switzerland

10 *ISO 13528:2005 (2005).* Statistical methods for use in proficiency testing by interlaboratory comparisons. Geneva, Switzerland

11 Analytical Methods Committee, Robust Statistics - How Not to Reject Outliers, Analyst, (1989) 114: p. 1693-1697.

14 ANNEXES

ANNEX 1 – Announcement of the PT on the IRMM webpage

ANNEX 2 – Announcement via e-mail and invitation

ANNEX 3 – Announcement of material dispatch

ANNEX 4 – Documents sent to participants

ANNEX 5 – Technical specifications of the calibration solutions

ANNEX 6 – Homogeneity of the test material

ANNEX 7 – Questionnaire and method performance data

ANNEX 8 – Data reported by participants

ANNEX 1: Announcement of the PT on the IRMM webpage

EU-RL PT 822: 4 EU marker PAHs in chocolate and cocoa butter

Proficiency Test on the determination of 4 EU marker PAHs in chocolate and cocoa butter

The European Union Reference Laboratory for Pylycyclic Aromatic Hydrocarbons organises a proficiency test on the determination of 4 EU marker PAHs in cocoa products.

The objective of this study is to evaluate the capabilities of EU food control laboratories in the determination of 4 EU marker PAHs in chocolate and cocoa butter.

Only national reference laboratories (NRLs) for PAHs and EU official food control laboratories (OCLs) can participate in the study.

Participation is admitted to maximum 40 official food control laboratories, which will be accepted in the order of registration.

Participation is free of charge for NRLs for PAHs. The participation fee for other official food control laboratories, which do not have national reference laboratory status, is **EUR 250** (two hundred fifty) per registration! Participation fees are due with the delivery of the test samples.

Test material and analytes

The test materials are commercial chocolate and cocoa butter samples containing the target analytes (see Table 1). One amber glass screw cap vial containing about 30 g of chocolate powder and one amber glass vial containing about 30 g of cocoa butter will be sent to the participants begin of July. In addition participants will get an ampoule with a solution of target PAHs with disclosed analyte content, in which - depending on their preference - either acetonitrile or toluene, will allow the participants verifying instrument calibration against an independent standard. Results do not have to be reporter for the standard solution.

The measurands are the 4 EU marker PAHs as listed in Table 1.

Results have to be reported for the contents of the individual analytes as well as for the sum of the four PAHs. Additional the analyte contents of the chocolate sample have to be expressed both on product and on fat basis. All results have to be reported corrected for recovery, and have to accompanied by the respective measurement uncertainty.

Table 1: measurands

benz[a]anthracene (BAA)	benzo[a]pyrene (BAP)
benzo[b]fluoranthene (BFF)	chrysene (CHR)

General outline

Participants are requested to perform three independent analysis of each sample using a method of their choice. The analyses shall be performed on the same day.

Performance assessment:

The performance of the participants in the determination of PAHs in the two test samples will be rated by z-scores

The standard deviations for proficiency assessment will be derived: - for all four analytes from the fitness-for-purpose function given in Commission Regulation (EU) no 836/2011 assuming a value of 0.3 µ/kg for the limit of detection

- for the sum parameter by propagating the individual standard deviations for proficiency assessment of the four analytes applying the law of error propagation.

Registration

Registration shall be done via this link.

Schedule

Registration	Sample dispatch	Reporting of results	Report
22 June 2012	Begin of July 2012	deadline 14/09/2012	December 2012

Latest update 30 May, 2012

ANNEX 2: Announcement of the PT via e-mail and confirmation of participation

WENZL Thomas (JRC-GEEL)

From: JRC IRMM CRL PAH
Sent: 29 May 2012 15:25
To: JRC IRMM CRL PAH
Subject: ILC822- Proficiency test on the determination of PAHs in chocolate and cocoa butter

Dear Madame/Sir,

Registration for participation in the inter-laboratory comparison study organised by the EU-RL PAH on the determination of the 4 EU priority PAHs in chocolate and cocoa butter has been opened. Participation is mandatory and free of charge for **national reference laboratories** (NRLs) for PAHs.
In support to the NRLs, to facilitate fulfilling their tasks according to Regulation No 882/2004, EU **official food control laboratories** falling under the responsibility of the NRLs may participate in the study. **The participation fee for official food control laboratories is 250 Euro per participation.** For reasons of logistics maximum 40 EU official food control laboratories can be accepted.
The target analytes are the 4 EU marker PAHs benz[*a*]anthracene, benzo[*a*]pyrene, benzo[*b*]fluoranthene, and chrysene. Results have to be reported corrected for recovery and accompanied by the respective measurement uncertainty for the individual PAHs as well as for the sum of the four PAHs. Additionally results for the chocolate sample have to be expressed both on product and on fat basis.

Each participant will be provided with one amber glass screw cap bottle containing ~ 30 g of a commercial chocolate sample, an amber glass vial containing about 30 g of cocoa butter. An ampoule with a standard solution in either acetonitrile or toluene with disclosed content may be used for verification of instrument calibration. Information on the preferred solvent will be requested via a separate email.

Detailed information can be found (from 30 May 2012 on) on the EU-RL website:
http://irmm.jrc.ec.europa.eu/html/CRLs/crl_pah/interlaboratory_comparisons/index.htm

Timing:

Deadline for registration: 22 June 2012
Dispatch of samples: Begin of July 2012
Deadline for reporting of results: 14 September 2012

Registration procedure:

Participants shall register via this link:
https://irmm.jrc.ec.europa.eu/ilc/ilcRegistration.do?selComparison=822

Registrations have to be confirmed by submitting the signed and stamped registration form via fax to the EU-RL (+32 14 571 783).

Access of NRLs to performance data of official food control laboratories:
Two options:
1) *NRL enrols official food control laboratories* and covers participation fees: NRL submits to EU-RL list of participants including name and address of laboratory, and details of the contact person (name, address (no post box!), email and telephone number). The coverage of the participation fees has to be confirmed and details for invoicing (e.g. order number) have to be provided. It shall be made clear, that the full participation fee is payable upon dispatch of the test samples. In return the performance data of the respective official food control laboratories will be disclosed to the NRL.
2) The *official food control laboratory enrols itself* in the inter-laboratory comparison and covers the participation fee: The official food control laboratory shall register via this link:
https://irmm.jrc.ec.europa.eu/ilc/ilcRegistration.do?selComparison=822
The NRL will get access to performance data of the official food control laboratory only upon providing a letter of consent.

Distribution of information:

The NRLs are kindly requested to distribute this information to the official food control

laboratories under their responsibility, and to assist the EU-RL in identifying laboratories that are eligible to participate in the study.

In case you may wish clarification of open questions, please do not hesitate to contact the EU-RL team via: **JRC-IRMM-CRL-PAH@ec.europa.eu**

With best regards
Thomas Wenzl

European Union Reference Laboratory for Polycyclic Aromatic Hydrocarbons (EU-RL PAH)

European Commission
DG Joint Research Centre

Institute for Reference Materials and Measurements
Food Safety and Quality Unit

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http://irmm.jrc.ec.europa.eu/

The views expressed are purely those of the writer and may not in any circumstances be regarded stating an official position of the European Commission

23/11/2012

Confirmation of participation

WENZL Thomas (JRC-GEEL)

From: JRC IRMM CRL PAH
Sent: 27 June 2012 19:28
To: WENZL Thomas (JRC-GEEL)
Subject: Solvent preference:

Dear Madame/Sir,

Hereby we confirm your participation in the proficiency test on the determination of four EU marker PAHs in chocolate and cocoa butter.

For this proficiency test we will supply you with a standard solution of PAHs with to you disclosed content, which shall serve you to check your instrument calibration against an external reference.

Depending of your preference, **you may choose between acetonitrile and toluene as solvent of the standard solution**.

Please tell us your preference by **replying to this email, mentioning the chosen solvent in the subject line** or email body.

We would like to dispatch the test samples within a fortnight. May I therefore ask you to reply to this email by Monday 02 July at the latest?

With best regards
Thomas Wenzl

Dr. Thomas Wenzl
Operating Manager of the EU-RL PAH

European Commission
DG Joint Research Centre
Institute for Reference Materials and Measurements
Food Safety and Quality Unit

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thomas.wenzl@ec.europa.eu

http://irmm.jrc.ec.europa.eu/

The views expressed are purely those of the writer and may not in any circumstances be stating an official position of the European Commission

ANNEX 3: Announcement of material dispatch

WENZL Thomas (JRC-GEEL)

From: JRC IRMM CRL PAH
Sent: 17 July 2012 10:16
To: JRC IRMM CRL PAH
Cc: BITTERHOF Almut (SANCO)
Subject: Proficiency test on the determination of PAHs in chocolate and cocoa butter

Dear Madame/Sir,

The test samples for the proficiency test on the determination of four EU marker PAHs in chocolate and cocoa butter were dispatched yesterday.
You should expect receipt of the parcel within 72 hours at the latest.

Please check the completeness of the delivery and confirm it by filling and retourning the sample receipt form to us (by fax) You will find the form in an envelop in the parcel together with your participation key, the outline of the study and the specification of the standard solution.

Please contact us in case you do not receive the samples by end of this week.

Information on how to report analysis results will be provided in the next days.
Deadline for reporting of analysis results is 14 September 2012.

With best regards
Thomas Wenzl

European Union Reference Laboratory for Polycyclic Aromatic Hydrocarbons (EU-RL PAH)

European Commission
DG Joint Research Centre

Institute for Reference Materials and Measurements
Food Safety and Quality Unit

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The views expressed are purely those of the writer and may not in any circumstances be stating an official position of the European Commission

ANNEX 4: Documents sent to participants

OUTLINE

EUROPEAN COMMISSION
JOINT RESEARCH CENTRE

Institute for Reference Materials and Measurements (Geel)
European Union Reference Laboratory for
Polycyclic Aromatic Hydrocarbons

European Union Reference Laboratory
Polycyclic Aromatic Hydrocarbons

Geel, 12 July 2012

ILC-822

Tenth inter-laboratory comparison study organised by the EU-RL PAHs

Analysis of four EU marker PAHs in chocolate and cocoa butter

Dear Madame/Sir,

The inter-laboratory comparison study organised by the EU-RL PAHs on the determination of four EU marker PAHs in chocolate and cocoa butter starts with the dispatch of the samples.

The target analytes are the four EU marker PAHs (benzo[*a*]pyrene, benzo[*b*]fluoranthene, benz[*a*]anthracene, and chrysene) and the participants are requested to report results on all of them.

Each participant will be provided with two screw cap vials containing a portion of a commercial chocolate sample, one crimp cap vial containing a commercial cocoa butter sample, and a known standard solution in either toluene or acetonitrile for checking of the instrument calibration against an external reference.

The content of the two vials containing the chocolate sample shall be merged and homogenised prior to analysis!

The cocoa butter shall be melted and homogenised prior to analysis.

Retieseweg 111, B-2440 Geel - Belgium. Telephone: (32-14) 571 211. http://irmm.jrc.ec.europa.eu
Telephone: direct line (32-14) 571 320. Fax: (32-14) 571 783.

E-mail: jrc-irmm-crl-pah@ec.europa.eu

1. Outline of the study.

The participating laboratories shall apply for the analyses a method of their choice.

The laboratories shall report the results by **14 September 2012 at the latest** via a web-based interface: **http://irmm.jrc.ec.europa.eu/Pages/ilcReporting.aspx**. Your participation/password key (required for reporting of results) is shipped together with the test samples (in the same parcel).

The participants are requested to report for both the chocolate and cocoa butter sample the results obtained from three replicate analyses. They also have to report for each sample both a single content value per analyte ("final value"), and the sum of the contents of the four analytes, on which the performance of the laboratory will be assessed. The "final value" for the chocolate sample shall be reported both on product basis (µg/kg chocolate) and on fat basis (µg/kg fat), as the latter is required by Commission Regulation (EU) No 835/2011.

Results have to be reported corrected for recovery, and the results for proficiency assessment ("final values") have to be accompanied by the respective measurement uncertainty (also for the sum parameter).

Participants are also requested to report together with the results details of the applied analysis method and some method performance characteristics of the applied analysis method.

Test materials and analytes

1. One crimp cap vial, labelled as "EU-RL PAHs, Interlaboratory comparison 822, 4 PAHs in cocoa based products, Cocoa butter", containing about 25 g of a *commercial cocoa butter:* The concentration of the individual analytes is in the range of about 0.5 µg/kg to 10 µg/kg. The analyte content shall be determined in triplicate. The participants have to report to the EU-RL besides the individual results of the replicate analyses also one value, on which they would like their performance to be assessed. This value is called on the reporting webpage for reasons of simplicity "final value".
2. Two screw cap vials, labelled as "EU-RL PAHs, Interlaboratory comparison 822, 4 PAHs in cocoa based products, Chocolate", containing each about 30 g of a *commercial chocolate*: The concentration of the individual analytes is in the range of 0.5 µg/kg to 10 µg/kg. The analyte content shall be determined in triplicate. Results have to be reported both on product and on fat basis. The participants have to report to the EU-RL besides the individual results of the replicate analyses also one value, on which they would like their performance to be assessed. This value is called on the reporting webpage for reasons of simplicity "final value". **The content of both vials shall be merged and homogenized before analysis!** The test portion size per analysis shall be at least 5 grams.
3. Depending of your preference, one ampoule, labelled as " Interlaboratory comparison 822, 4 PAHs in cocoa based products ACN-K-822", or " Interlaboratory comparison 822, 4 PAHs in cocoa based products TOL-K-822" with about 3 ml of a solution of *15+1 EU priority* ***PAHs in acetonitrile(ACN-K-822) respectively toluene (TOL-K-822)***. The analyte concentration of your preferred solution is given in the attached document. The solutions may be used by the participants to check their instrument calibration against an independent reference. Participants do not have to report results for this solution.

Please bear in mind that the solutions *do not contain any internal standard*. The standard solution in acetonitrile contains small amounts of toluene, which stem from the preparation of stock solution from neat materials.

The analyte composition of the standard solutions is given in Table 1 (*please note the acronyms for reporting*):

Table 1: PAHs contained in the standard solutions

benz[*a*]anthracene (BAA)	**benzo[*a*]pyrene (BAP)**
benzo[*b*]fluoranthene (BBF)	**chrysene (CHR)**
benzo[*j*]fluoranthene	cyclopenta[*cd*]pyrene
benzo[*k*]fluoranthene	dibenz[*a,h*]anthracene
benzo[*c*]fluorene	dibenzo[*a,e*]pyrene
benzo[*ghi*]perylene	dibenzo[*a,h*]pyrene
dibenzo[*a,i*]pyrene	dibenzo[*a,l*]pyrene
indeno[1,2,3-*cd*]pyrene	5-methylchrysene

The four target PAHs are given in bold.

Contact person

Thomas Wenzl

Institute for Reference Materials and Measurements (IRMM)
Retieseweg 111
B-2440 Geel, Belgium
Tel: +32-14-571 320
FAX: +32-14-571 783

E-mail: jrc-irmm-crl-pah@ec.europa.eu

In case of questions please do not hesitate to contact us.

With kind regards,

Thomas Wenzl

(Operating Manager of the European Union Reference Laboratory for Polycyclic Aromatic Hydrocarbons)

Cc: Almut Bitterhof, Franz Ulberth, Stefanka Bratinova

SAMPLE RECEIPT

EUROPEAN COMMISSION
JOINT RESEARCH CENTRE

Institute for Reference Materials and Measurements (Geel)
European Union Reference Laboratory for
Polycyclic Aromatic Hydrocarbons

European Union Reference Laboratory
Polycyclic Aromatic Hydrocarbons

Geel, 27.07.2012

ILC 822

Tenth inter-laboratory comparison study organised by the EU-RL PAHs

Analysis of four EU marker PAHs in chocolate and cocoa butter

Confirmation of the receipt of the samples

RECEIPT FORM

Surname of Participant	
Name of Participant	
Affiliation	
Password key	
Country	

Content of the parcel

a) Two screw cap bottles containing the chocolate test material

b) Two crimp cap bottle containing the cocoa butter test material

c) One inter-laboratory comparison sample receipt form (= this form)

Please ensure that the items listed below have been received undamaged, and then describe the relevant statement:

Date of the receipt of the test materials	
All items have been received undamaged	YES ☐ / NO ☐
If NO, please list damaged items according to the letters associated at each item in the list above (in case of samples,	

Retieseweg 111, B-2440 Geel - Belgium. Telephone: (32-14) 571 211. http://irmm.jrc.ec.europa.eu
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E-mail: jrc-irmm-crl-pah@ec.europa.eu

please specify the code too) Please write one item per row	
Items are missing	YES ☐ / NO ☐
If YES, please list missing items according to the letters associated at each item in the list above Please write one item per row	
Serial number of the chocolate samples you received Please write one item per row	
Serial number of the cocoa butter sample you received	
Ampoule number of the standard solution	

Signature .

Store the chocolate and cocoa butter samples at room temperature in the dark

ATTENTION

Please, submit the filled form by fax to the attention of Thomas Wenzl to the following number:

+32 – (0)14 - 571 783

EUROPEAN COMMISSION
JOINT RESEARCH CENTRE

Institute for Reference Materials and Measurements (Geel)
European Union Reference Laboratory for Polycyclic Aromatic Hydrocarbons

European Union Reference Laboratory
Polycyclic Aromatic Hydrocarbons

Geel, 18.07.2012

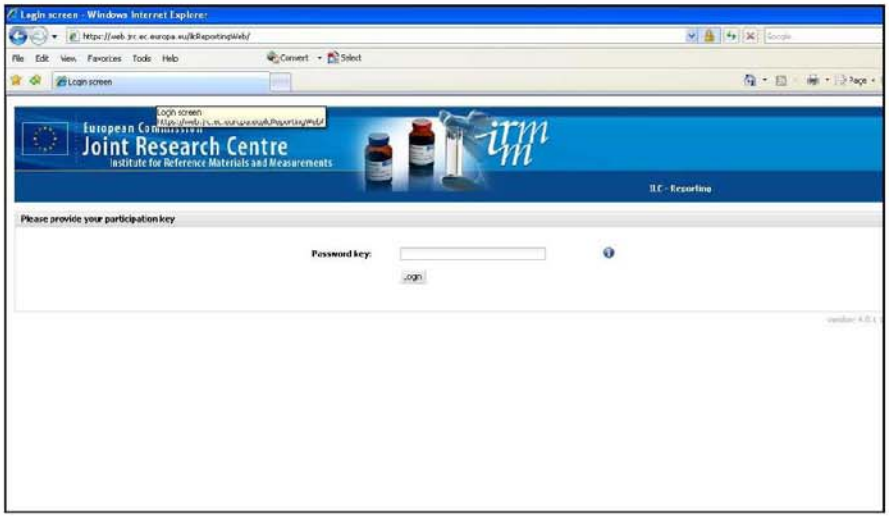
ILC 822

Proficiency test on the determination of four EU marker PAHs in chocolate and cocoa butter

Guidance for reporting of analysis results

Please enter the participation/password key to access the reporting webpage.

The participation/password key was sent to you together with the samples.



After the login you will get to the start page (Figure 2).

It provides different reporting options. You can report data sorted according to the analyte or according to the test sample. You can also report data for all samples/analytes at once. You may select the option which suits you best. There is also the possibility to download an Excel template, to fill it offline and to submit it at a later date online (Figure 3). Instructions on how to do this are given on the download page.

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Telephone: direct line (32-14) 571 320. Fax: (32-14) 571 783.

E-mail: jrc-irmm-crl-pah@ec.europa.eu

Figure 2: Start page

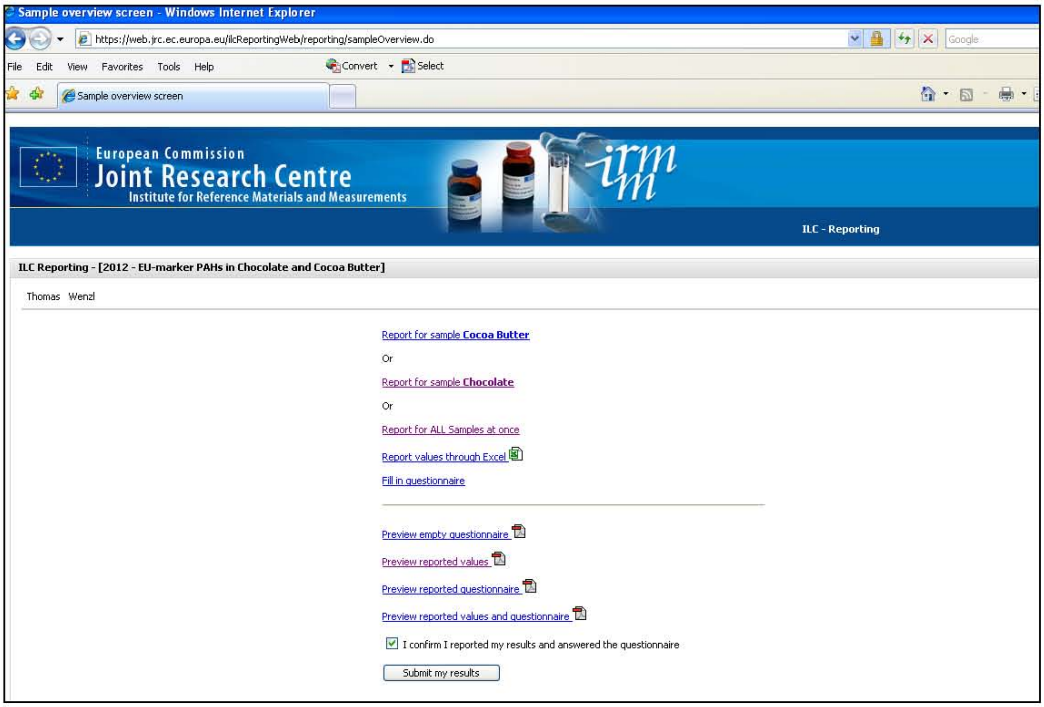


Figure 4: Excel download

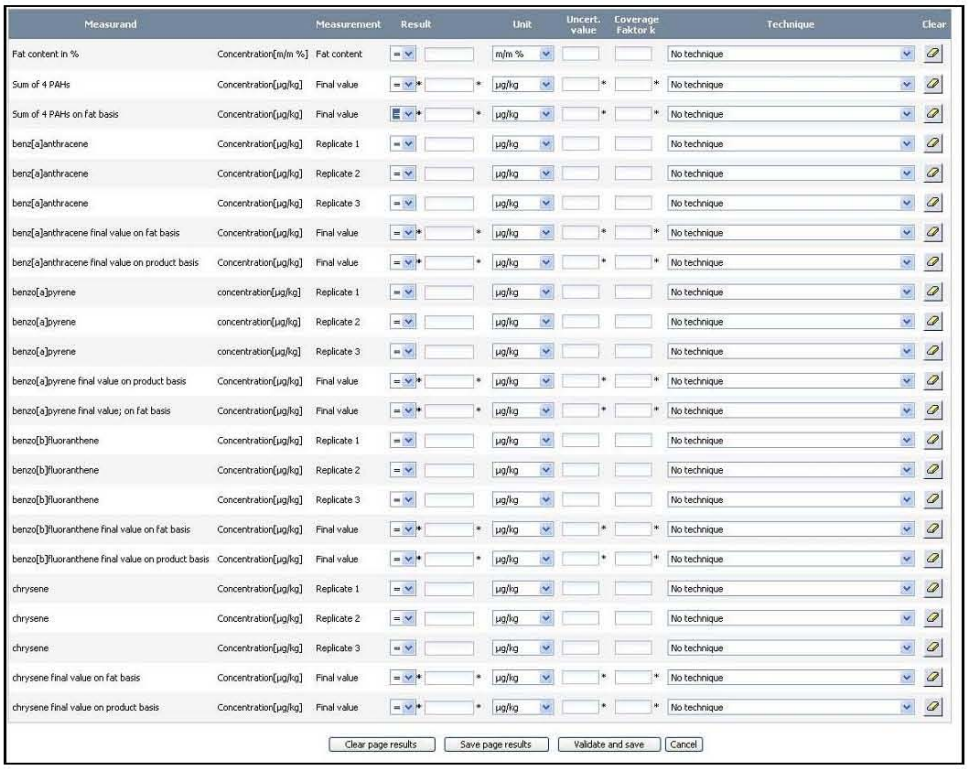
The reporting page for the chocolate sample is given in Figure 4. The page for the cocoa sample looks similar. Besides the results of individual replicate measurements a "final value" is requested, on which the performance will be assessed. The sum of the four

PAHs has to be reported too. For the chocolate sample, the fat content of the sample has to be reported and the results have to be expressed both on chocolate and on fat basis.

Fields indicated with an asterisk are mandatory!

If the result of a measurement was **below LOD/LOQ**, please select the "<" sign and report the respective numerical value of LOD/LOQ. Only numerical values, with a dot as comma separator, may be put into the results field.

Figure 4: Reporting page for chocolate sample



Measurand		Measurement	Result	Unit	Uncert. value	Coverage Faktor k	Technique	Clear
Fat content in %	Concentration[m/m %]	Fat content	=	m/m %			No technique	
Sum of 4 PAHs	Concentration[µg/kg]	Final value	= *	µg/kg	*	*	No technique	
Sum of 4 PAHs on fat basis	Concentration[µg/kg]	Final value	= *	µg/kg	*	*	No technique	
benz[a]anthracene	Concentration[µg/kg]	Replicate 1	=	µg/kg			No technique	
benz[a]anthracene	Concentration[µg/kg]	Replicate 2	=	µg/kg			No technique	
benz[a]anthracene	Concentration[µg/kg]	Replicate 3	=	µg/kg			No technique	
benz[a]anthracene final value on fat basis	Concentration[µg/kg]	Final value	= *	µg/kg	*	*	No technique	
benz[a]anthracene final value on product basis	Concentration[µg/kg]	Final value	= *	µg/kg	*	*	No technique	
benzo[a]pyrene	concentration[µg/kg]	Replicate 1	=	µg/kg			No technique	
benzo[a]pyrene	concentration[µg/kg]	Replicate 2	=	µg/kg			No technique	
benzo[a]pyrene	concentration[µg/kg]	Replicate 3	=	µg/kg			No technique	
benzo[a]pyrene final value on product basis	Concentration[µg/kg]	Final value	= *	µg/kg	*	*	No technique	
benzo[a]pyrene final value; on fat basis	Concentration[µg/kg]	Final value	= *	µg/kg	*	*	No technique	
benzo[b]fluoranthene	Concentration[µg/kg]	Replicate 1	=	µg/kg			No technique	
benzo[b]fluoranthene	Concentration[µg/kg]	Replicate 2	=	µg/kg			No technique	
benzo[b]fluoranthene	Concentration[µg/kg]	Replicate 3	=	µg/kg			No technique	
benzo[b]fluoranthene final value on fat basis	Concentration[µg/kg]	Final value	= *	µg/kg	*	*	No technique	
benzo[b]fluoranthene final value on product basis	Concentration[µg/kg]	Final value	= *	µg/kg	*	*	No technique	
chrysene	Concentration[µg/kg]	Replicate 1	=	µg/kg			No technique	
chrysene	Concentration[µg/kg]	Replicate 2	=	µg/kg			No technique	
chrysene	Concentration[µg/kg]	Replicate 3	=	µg/kg			No technique	
chrysene final value on fat basis	Concentration[µg/kg]	Final value	= *	µg/kg	*	*	No technique	
chrysene final value on product basis	Concentration[µg/kg]	Final value	= *	µg/kg	*	*	No technique	

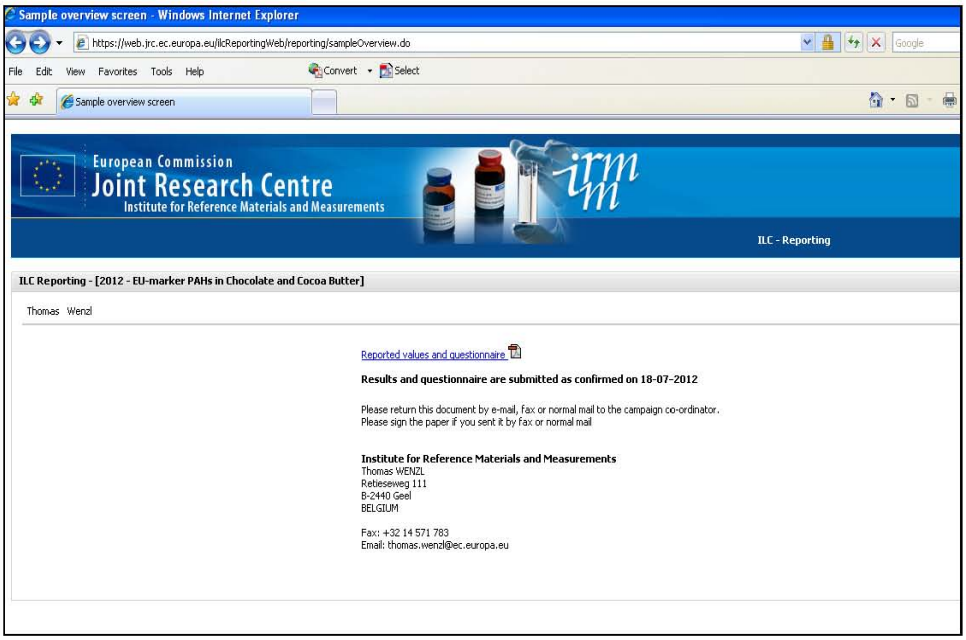
Clear page results | Save page results | Validate and save | Cancel

Please save the entered results for each page ("save page results") or "validate and save" them. Neither saving nor validating and saving will submit the data to the organisers of the PT. This has to be done separately and includes the review and approval of your data (see Figure 2). Please fill also the questionnaire on details of the applied analysis method (see Figure 2). We advice you to preview the empty questionnaire, in order to collect all required information prior to reporting of results.

The reporting system however will evaluate if all mandatory data (analysis results and information in the questionnaire) was provided.

You may preview the reported data by applying the respective preview links. Once you submit the results ("submit my results" button) you cannot modify them anymore. After submission of the results, you will be directed to a page on which you will find a pdf file containing your reported data (see Figure 5). Please send this file or a copy of it to the organisers of the PT.

Figure 5: Confirmation of data reporting



A bug in the system requests unfortunately reporting of uncertainty and coverage factor values also for results that were not set mandatory. If you do not want to report this data (for Replicate 1 to Replicate 3) then put just 99 into the fields for uncertainty and coverage factor. We will erase them later from the database.

Our informatics department is already busy with fixing the bug. However, the updated version will be released begin of August at the earliest.

We would like to apologize for any inconvenience this might cause.

With best regards

Thomas Wenzl

PARTICIPANT CODES

EUROPEAN COMMISSION
JOINT RESEARCH CENTRE

Institute for Reference Materials and Measurements (Geel)
European Union Reference Laboratory for Polycyclic Aromatic Hydrocarbons

EURL
European Union Reference Laboratory
Polycyclic Aromatic Hydrocarbons

Geel, 05.07.2012

Dear «Title» «Firstname» «Surname»
«Organisation» - «Department»
«Address»
«Zip» - «Town»
«Country»
«LName»

Dear Madame/Sir

Please find below your participation key. You need this unique key for the reporting of results via the web portal: **http://irmm.jrc.ec.europa.eu/Pages/ilcReporting.aspx**

Participation/password key: ……

Your laboratory code is: ….

Results have to be reported before 14 September 2012!

With best regards

Thomas Wenzl

(Operating manager of the EU-RL PAHs)

Retieseweg 111, B-2440 Geel - Belgium. Telephone: (32-14) 571 211. http://irmm.jrc.ec.europa.eu
Telephone: direct line (32-14) 571 320. Fax: (32-14) 571 783.

E-mail: jrc-irmm-crl-pah@ec.europa.eu

ANNEX 5: Technical specifications of the calibration solutions

ACETONITRILE SOLUTION

EUROPEAN COMMISSION
JOINT RESEARCH CENTRE

Institute for Reference Materials and Measurements (Geel)
European Union Reference Laboratory for Polycyclic Aromatic Hydrocarbons

European Union Reference Laboratory
Polycyclic Aromatic Hydrocarbons

Standard solution specification sheet	**Product ID: ACN-K–822**
Date of production: 10/08/2011	Total volume: 3 mL
Expiry date: *September 2012*	

Standard solution composition:

	Product name	CAS	Conc.*	Conc.*	U**
			(ng/g)	(ng/ml)	± %
1	5-methylchrysene	3697-24-3	64.2	50.1	1
2	Benz[*a*]anthracene	56-55-3	64.1	50.0	1
3	Benzo[*a*]pyrene	50-32-8	64.2	50.1	1
4	Benzo[*b*]fluoranthene	205-99-2	65.4	51.1	1
5	Benzo[*c*]fluorene	205-12-9	63.3	49.4	1
6	Benzo[*ghi*]perylene	191-24-2	65.5	51.2	1
7	Benzo[*j*]fluoranthene	205-82-3	64.8	50.6	1
8	Benzo[*k*]fluoranthene	207-08-9	64.9	50.7	1
9	Chrysene	218-01-9	64.3	50.2	1
10	Cyclopenta[*cd*]pyrene	27208-37-3	67.8	52.9	2
11	Dibenzo[*a,e*]pyrene	192-65-4	64.3	50.2	1
12	Dibenz[*a,h*]anthracene	53-70-3	64.3	50.2	1
13	Dibenzo[*a,h*]pyrene	189-64-0	64.6	50.4	1
14	Dibenzo[*a,i*]pyrene	189-55-9	65.6	51.2	3
15	Dibenzo[*a,l*]pyrene	191-30-0	64.8	50.6	1
16	Indeno[1,2,3-*cd*]pyrene	193-39-5	63.5	49.6	1

** The concentrations were calculated taking into account the purity statements of the single products. The concentration value given in ng/mL is based on the gravimetrical preparation data and the nominal volume of the applied volumetric flask.*

*** U is the expanded uncertainty calculated using the coverage factor 2 (corresponding to a confidence interval of 95%) multiplied by the combined standard uncertainty. The standard uncertainty is equal to the square root of the sum of the squares of the uncertainties associated with each single operation involved in the preparation of this standard solution.*

Solvent: Acetonitrile

Retieseweg 111, B-2440 Geel - Belgium. Telephone: (32-14) 571 211. http://irmm.jrc.ec.europa.eu
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E-mail: jrc-irmm-crl-pah@ec.europa.eu

TOLUENE SOLUTION

EUROPEAN COMMISSION
JOINT RESEARCH CENTRE

Institute for Reference Materials and Measurements (Geel)
European Union Reference Laboratory for Polycyclic Aromatic Hydrocarbons

European Union Reference Laboratory
Polycyclic Aromatic Hydrocarbons

Standard solution specification sheet	**Product ID: TOL-K–822**
Date of production: 10/08/2011	Total volume: 3 mL
Expiry date: *September 2012*	

Standard solution composition:

	Product name	CAS	Conc.*	Conc.*	U**
			(ng/g)	(ng/ml)	± %
1	5-methylchrysene	3697-24-3	57.9	50.1	1
2	Benz[*a*]anthracene	56-55-3	57.9	50.1	1
3	Benzo[*a*]pyrene	50-32-8	58.0	50.1	1
4	Benzo[*b*]fluoranthene	205-99-2	59.0	51.0	1
5	Benzo[*c*]fluorene	205-12-9	57.2	49.4	1
6	Benzo[*ghi*]perylene	191-24-2	59.2	51.2	1
7	Benzo[*j*]fluoranthene	205-82-3	58.4	50.5	1
8	Benzo[*k*]fluoranthene	207-08-9	58.5	50.6	1
9	Chrysene	218-01-9	58.3	50.4	1
10	Cyclopenta[*cd*]pyrene	27208-37-3	60.8	52.5	2
11	Dibenzo[*a,e*]pyrene	192-65-4	57.9	50.1	1
12	Dibenz[*a,h*]anthracene	53-70-3	59.1	51.1	1
13	Dibenzo[*a,h*]pyrene	189-64-0	58.1	50.2	1
14	Dibenzo[*a,i*]pyrene	189-55-9	59.4	51.4	3
15	Dibenzo[*a,l*]pyrene	191-30-0	58.6	50.7	1
16	Indeno[1,2,3-*cd*]pyrene	193-39-5	57.3	49.6	1

** The concentrations were calculated taking into account the purity statements of the single products. The concentration value given in ng/mL is based on the gravimetrical preparation data and the nominal volume of the applied volumetric flask.*

*** U is the expanded uncertainty calculated using the coverage factor 2 (corresponding to a confidence interval of 95%) multiplied by the combined standard uncertainty. The standard uncertainty is equal to the square root of the sum of the squares of the uncertainties associated with each single operation involved in the preparation of this standard solution.*

Solvent: Toluene

Retieseweg 111, B-2440 Geel - Belgium. Telephone: (32-14) 571 211. http://irmm.jrc.ec.europa.eu
Telephone: direct line (32-14) 571 320. Fax: (32-14) 571 783.

E-mail: jrc-irmm-crl-pah@ec.europa.eu

ANNEX 6: Homogeneity of the test material

COCOA BUTTER TEST MATERIAL

PRO - 23/11/2012 BaA

Analyte: **BAA** Matrix: **Cocoa butter**

	n =	9		
	mean =	1.466667	22%	**= σ-trg(%)**
0.000625	**s_x =**	0.025	0.322667	**= σ-trg**
√MSW =	**s_w =**	0.057735		
	s_s =	0.032275	0.0968	**= 0,3*σ**

ISO-13528 passed

F = 0.375 3.020383 = Fcrit

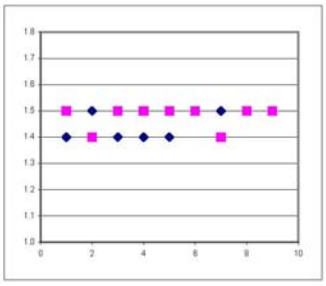
passed

IUPAC

(MSB-MSW)/2 -0.00104 0.020983 = $F1*(0,3*\sigma)^2+F2*MSW$

passed

Bottle	Result a	Result b	diff	sum	avg
Iddentification Code: 007	1.4	1.5	-0.1	2.9	1.45
Iddentification Code: 016	1.5	1.4	0.1	2.9	1.45
Iddentification Code: 017	1.4	1.5	-0.1	2.9	1.45
Iddentification Code: 021	1.4	1.5	-0.1	2.9	1.45
Iddentification Code: 058	1.4	1.5	-0.1	2.9	1.45
Iddentification Code: 061	1.5	1.5	0	3	1.5
Iddentification Code: 102	1.5	1.4	0.1	2.9	1.45
Iddentification Code: 111	1.5	1.5	0	3	1.5
Iddentification Code: 125	1.5	1.5	0	3	1.5



$\Sigma(diff)^2$ = 0.06

var(sum)/2 = 0.00125 =MSB

PRO - 23/11/2012 CHR

Analyte: **CHR** Matrix: **Cocoa butter**

	n =	9		
	mean =	1.861111	22%	**= σ-trg(%)**
0.004236111	**s_x =**	0.065085	0.409444	**= σ-trg**
√MSW =	**s_w =**	0.113039		
	s_s =	0.046398	0.122833	**= 0,3*σ**

ISO-13528 passed

F = 0.663043 3.020383 = Fcrit

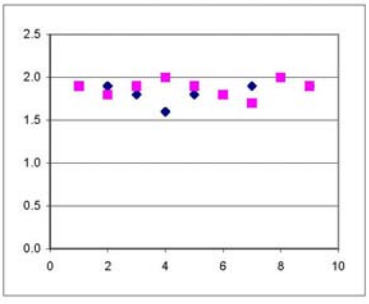
passed

IUPAC

(MSB-MSW)/2 -0.00215 0.041271 = $F1*(0,3*\sigma)^2+F2*MSW$

passed

Bottle	Result a	Result b	diff	sum	avg
Iddentification Code: 007	1.9	1.9	0	3.8	1.9
Iddentification Code: 016	1.9	1.8	0.1	3.7	1.85
Iddentification Code: 017	1.8	1.9	-0.1	3.7	1.85
Iddentification Code: 021	1.6	2	-0.4	3.6	1.8
Iddentification Code: 058	1.8	1.9	-0.1	3.7	1.85
Iddentification Code: 061	1.8	1.8	0	3.6	1.8
Iddentification Code: 102	1.9	1.7	0.2	3.6	1.8
Iddentification Code: 111	2	2	0	4	2
Iddentification Code: 125	1.9	1.9	0	3.8	1.9



$\Sigma(diff)^2$ = 0.23

var(sum)/2 = 0.008472 =MSB

PRO - 23/11/2012 BbF

Analyte: **BBF** Matrix: **Cocoa butter**

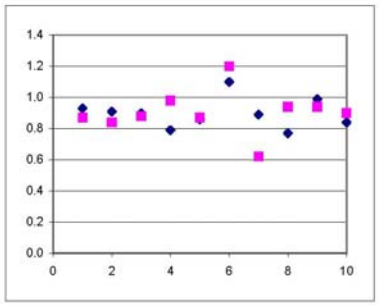
	n =	10		
	mean =	0.901	22%	**= σ-trg(%)**
0.010316	**s_x =**	0.101566	0.19822	**= σ-trg**
√MSW =	**s_w =**	0.090277		
	s_s =	0.078997	0.059466	**= 0,3*σ**

ISO-13528 failed

F = 2.531425 3.020383 = Fcrit
passed

IUPAC
(MSB-MSW)/2 0.006241 0.01488 = F1*(0,3*σ)^2+F2*MSW
passed

Bottle	Result a	Result b	diff	sum	avg
Identification	0.9	0.87	0.06	1.8	0.9
Identification	0.9	0.84	0.07	1.75	0.875
Identification	0.9	0.88	0.02	1.78	0.89
Identification	0.8	0.98	-0.19	1.77	0.885
Identification	0.9	0.87	-0.01	1.73	0.865
Identification	1.1	1.2	-0.1	2.3	1.15
Identification	0.9	0.62	0.27	1.51	0.755
Identification	0.8	0.94	-0.17	1.71	0.855
Identification	0.99	0.94	0.05	1.93	0.965
Identification	0.84	0.9	-0.06	1.74	0.87



Σ(diff)^2 = 0.163
var(sum)/2 = 0.020631 =MSB

PRO - 23/11/2012 BaP

Analyte: **BAP** Matrix: **Cocoa butter**

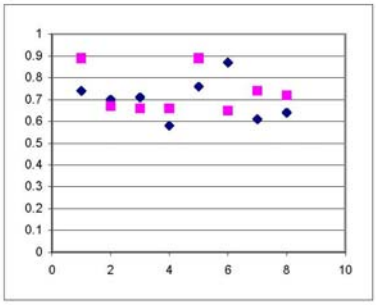
	n =	8		
	mean =	0.718125	22%	**= σ-trg(%)**
0.005385268	**s_x =**	0.073384	0.157988	**= σ-trg**
√MSW =	**s_w =**	0.086927		
	s_s =	0.040089	0.047396	**= 0,3*σ**

ISO-13528 passed

F = 1.425381 3.020383 = Fcrit
passed

IUPAC
(MSB-MSW)/2 0.001607 0.011855 = F1*(0,3*σ)^2+F2*MSW
passed

Bottle	Result a	Result b	diff	sum	avg
Identification Code: 007	0.74	0.89	-0.15	1.63	0.815
Identification Code: 016	0.7	0.67	0.03	1.37	0.685
Identification Code: 017	0.71	0.66	0.05	1.37	0.685
Identification Code: 058	0.58	0.66	-0.08	1.24	0.62
Identification Code: 061	0.76	0.89	-0.13	1.65	0.825
Identification Code: 102	0.87	0.65	0.22	1.52	0.76
Identification Code: 111	0.61	0.74	-0.13	1.35	0.675
Identification Code: 125	0.64	0.72	-0.08	1.36	0.68



Σ(diff)^2 = 0.1209
var(sum)/2 = 0.010771 =MSB

CHOCOLATE TEST MATERIAL

PRO - 23/11/2012 BaA_for report

Analyte: **BAA** Matrix: **Chocolate**

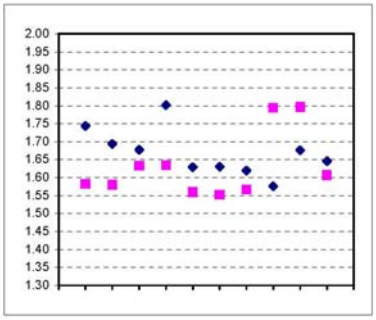
	n =	10		
	mean =	1.6501	22%	**= σ-trg(%)**
0.002655095	**s_x =**	0.0515	0.3630	**= σ-trg**
ÖMSW =	**s_w =**	0.0855		
	s_s =	0.0316	0.1089	**= 0,3*s**

ISO-13528 passed

F = 0.7261477 3.02038295 = Fcrit
passed

IUPAC
(MSB-MSW)/2 -0.0010 0.0297 = F1*(0,3*s)²+F2*MSW
passed

Bottle	Result a	Result b	diff	sum	avg
Ampoule 5	1.74	1.58	0.16165736	3.32609376	1.66304688
Ampoule 37	1.69	1.58	0.11375964	3.27468967	1.63734484
Ampoule 43	1.68	1.63	0.04428027	3.31099415	1.65549708
Ampoule 67	1.80	1.63	0.16717621	3.43641899	1.7182095
Ampoule 82	1.63	1.56	0.06939947	3.18853231	1.59426615
Ampoule 99	1.63	1.55	0.07796052	3.18409098	1.59204549
Ampoule 112	1.62	1.57	0.05232577	3.18672079	1.5933604
Ampoule 121	1.58	1.79	-0.21825556	3.36985831	1.68492916
Ampoule 152	1.68	1.80	-0.12040092	3.47247039	1.73623519
Ampoule 159	1.65	1.61	0.03885248	3.25295349	1.62647674



∑(diff)² = 0.14625649
var(sum)/2 = 0.00531 =MSB

PRO - 23/11/2012 BaP_for report

Analyte: **BAP** Matrix: **Chocolate**

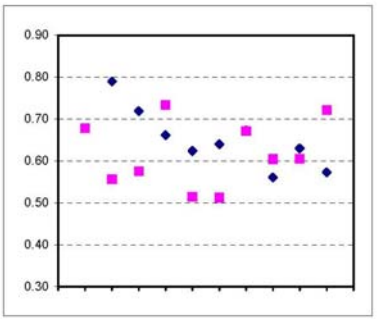
	n =	10		
	mean =	0.6602	22%	**= σ-trg(%)**
0.010313776	**s_x =**	0.1016	0.1452	**= σ-trg**
ÖMSW =	**s_w =**	0.1357		
	s_s =	0.0333	0.0436	**= 0,3*s**

ISO-13528 passed

F = 1.12085077 3.02038295 = Fcrit
passed

IUPAC
(MSB-MSW)/2 0.0011 0.0222 = F1*(0,3*s)²+F2*MSW
passed

Bottle	Result a	Result b	diff	sum	avg
Ampoule 5	1.16	0.68	0.48448626	1.84002522	0.92001261
Ampoule 37	0.79	0.56	0.23346367	1.34600795	0.67300398
Ampoule 43	0.72	0.57	0.14395893	1.29373568	0.64686784
Ampoule 67	0.66	0.73	-0.0720731	1.39520191	0.69760096
Ampoule 82	0.62	0.51	0.10965853	1.13803986	0.56901993
Ampoule 99	0.64	0.51	0.12782347	1.15191791	0.57595896
Ampoule 112	0.67	0.67	0.00146784	1.34413807	0.67206903
Ampoule 121	0.56	0.60	-0.04386024	1.1649992	0.5824996
Ampoule 152	0.63	0.61	0.02462129	1.23508355	0.61754178
Ampoule 159	0.57	0.72	-0.14840052	1.29412621	0.64706311



∑(diff)² = 0.36806956
var(sum)/2 = 0.02063 =MSB

PRO - 23/11/2012 BbF_for report

Analyte: **BBF** Matrix: **Chocolate**

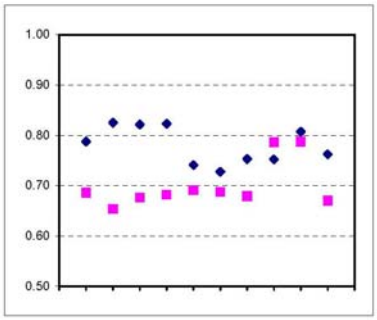
	n =	10		
	mean =	0.7400	22%	**= σ-trg(%)**
0.000791281	**s_x =**	0.0281	0.1628	**= σ-trg**
ÖMSW =	**s_w =**	0.0709		
	s_s =	0.0415	0.0488	**= 0,3*s**

ISO-13528 passed

F = 0.31445923 3.02038295 = Fcrit
passed

IUPAC
(MSB-MSW)/2 -0.0017 0.0096 = F1*(0,3*s)^2+F2*MSW
passed

Bottle	Result a	Result b	diff	sum	avg
Ampoule 5	0.79	0.69	0.1022811	1.47365823	0.73682911
Ampoule 37	0.83	0.65	0.17106593	1.47915218	0.73957609
Ampoule 43	0.82	0.68	0.14580566	1.49766839	0.74883419
Ampoule 67	0.82	0.68	0.14153493	1.50471558	0.75235779
Ampoule 82	0.74	0.69	0.050544	1.43212121	0.71606061
Ampoule 99	0.73	0.69	0.03965039	1.41546838	0.70773419
Ampoule 112	0.75	0.68	0.0737462	1.43210425	0.71605212
Ampoule 121	0.75	0.79	-0.0343326	1.53813427	0.76906713
Ampoule 152	0.81	0.79	0.01999404	1.59454582	0.79727291
Ampoule 159	0.76	0.67	0.0921558	1.43234391	0.71617196



$\sum$(diff)^2 = 0.10065294
var(sum)/2 = 0.00158 =MSB

PRO - 23/11/2012 CHR_for report

Analyte: **CHR** Matrix: **Chocolate**

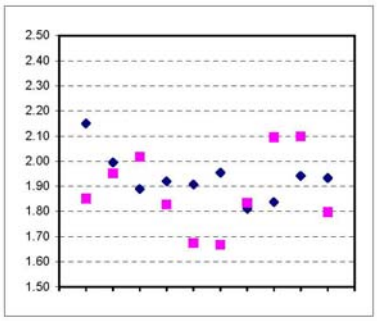
	n =	10		
	mean =	1.9078	22%	**= σ-trg(%)**
0.007135684	**s_x =**	0.0845	0.4197	**= σ-trg**
ÖMSW =	**s_w =**	0.1347		
	s_s =	0.0440	0.1259	**= 0,3*s**

ISO-13528 passed

F = 0.7869466 3.02038295 = Fcrit
passed

IUPAC
(MSB-MSW)/2 -0.0019 0.0481 = F1*(0,3*s)^2+F2*MSW
passed

Bottle	Result a	Result b	diff	sum	avg
Ampoule 5	2.15	1.85	0.29869719	4.00216626	2.00108313
Ampoule 37	1.99	1.95	0.04229567	3.94719986	1.97359993
Ampoule 43	1.89	2.02	-0.1290009	3.90658502	1.95329251
Ampoule 67	1.92	1.83	0.09174923	3.74750676	1.87375338
Ampoule 82	1.91	1.67	0.23296986	3.58247701	1.79123851
Ampoule 99	1.95	1.67	0.28683474	3.62237917	1.81118958
Ampoule 112	1.81	1.83	-0.02312572	3.6451847	1.82259235
Ampoule 121	1.84	2.10	-0.25793443	3.93248074	1.96624037
Ampoule 152	1.94	2.10	-0.15629393	4.03967726	2.01983863
Ampoule 159	1.93	1.80	0.13635385	3.73045032	1.86522516



$\sum$(diff)^2 = 0.36270235
var(sum)/2 = 0.01427 =MSB

ANNEX 7: Questionnaire and method performance characteristics

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Milc questionnaire

Comparison for 2012 - EU-marker PAHs in Chocolate and Cocoa Butter

Please report the method performance parameters for the determination of PAHs in the chocolate and cocoa butter material as indicated below. The unit for limit of detection (LOD), limit of quantitation (LOQ) is µg/kg. The method recovery shall be reported as percentage (%) and has to be understood as the yield of the method. Please describe also the key elements of the applied analysis procedure. Thank you for your cooperation. The EU-RL Team

Submission Form

1. **Is the applied analysis method accredited according to ISO 17025?**
- ○ a) Yes
- ○ b) No

2. **Are chocolate/cocoa butter within the scope of the accredited method?**
- ○ a) Yes
- ○ b) No

3. **How many chocolate samples did you analyse so far for PAH content?**
- ○ A) <10
- ○ B) 10-50
- ○ C) 50-100
- ○ D) >100

4. **Which analysis technique did you apply?**
- ☐ A) GC-FID
- ☐ B) GC-MS
- ☐ C) GC-MS/MS
- ☐ D) GC-HRMS
- ☐ E) HPLC-FD
- ☐ F) HPLC-UV/FD
- ☐ G) LC-MS
- ☐ H) LC-MS/MS
- ☐ I) UHPLC-FD
- ☐ J) UHPLC-UV/FD

5. **Which chromatographic column did you apply for the analyses?**

6. **Which sample amount did you take per analysis? (g)**
(number)

7. **Which of the following sample preparation procedures did you apply for the chocolate sample?** *
- ☐ 1) Extraction with organic solvent
- ☐ 2) Liquid/Liquid partitioning
- ☐ 3) Saponification
- ☐ 4) Chromatography/fractionation

7.1. If applicable: Which extraction technique was applied?
- ☐ A) Pressurised liquid extraction (PLE)
- ☐ B) Sonication
- ☐ C) Soxhlet extraction
- ☐ D) Other

7.2. If applicable: Which chromatography/fractionation technique was applied?
- ☐ A) Column chromatography on silica
- ☐ B) Gel permeation chromatography (GPC)
- ☐ C) Donor acceptor complex chromatography (DACC)
- ☐ D) Solid phase extraction (SPE)
- ☐ E) Other

7.2.1. If applicable: Please specify SPE cartridge(s).

8. **How did you calibrate your instrument?**

8.1. Did you apply external calibration? *
- ○ a) Yes
- ○ b) No

8.1.1. In case of external calibration: How did you calibrate? *
- ☐ A) with standards in an organic solvent
- ☐ B) with matrix matched standards

8.2. Did you apply internal standardisation? *

- ○ a) Yes
- ○ b) No

8.2.1. Which internal standards did you apply *

- ☐ A) Structural analogue(s) of the analyte(s)
- ☐ B) Stable isotope labelled analogue(s)

8.2.2. Please provide details on the applied internal standards *

8.3. Did you apply standard addition? *

- ○ a) Yes
- ○ b) No

9. **How did you obtain the PAH contents of the chocolate test material expressed on fat basis?**

- ☐ A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.
- ☐ B) Extracting fat from the chocolate matrix and performing analysis on the fat fraction

10. **How did you determine the fat content of the chocolate sample?** *

- ☐ A) Hydrolysis of the sample followed by solvent extraction
- ☐ B) Only solvent extraction
- ☐ C) Near infrared spectroscopy (NIR)
- ☐ D) Other

11. **Did you experience any problems during sample preparation of the chocolate/coco butter sample?**

- ○ a) Yes
- ○ b) No

11.1. Please specify: *

12. **Did you experience chromatographic interferences?**

- ○ a) Yes
- ○ b) No

12.1. Please specify: *

13. **Please provide details of method performance parameters for the determination of PAHs in chocolate** *

See table **Table 1: chocolate** at bottom

14. **Please provide details on method performance parameters for the determination of PAHs in cocoa butter** *

See table **Table 2: cocoa butter** at bottom

Table 1: chocolate

LOD and LOQ shall be reported in µg/kg chocolate. Recovery shall be given in %, and has to be understood as yield.

Questions/Response table	*LOD*	*LOQ*	*Recovery*
BAA			
BAP			
BBF			
CHR			

Table 2: cocoa butter

LOD and LOQ shall be expressed in µg/kg cocoa butter. Recovery shall be given in %, and has to be understood as yield.

Questions/Response table	*LOD*	*LOQ*	*Recovery*
BAA			
BAP			
BBF			
CHR			

QUESTIONNAIRE:

1) Is the applied analysis method accredited according to ISO 17025?
2) Are chocolate/cocoa butter within the scope of the accredited method?
3) How many chocolate samples did you analyse so far for PAH content?
4) Which analysis technique did you apply
5) Which chromatographic column did you apply for the analyses?
6) Which sample amount did you take per analysis?

Participant	1: Accreditation	2: within Scope	3:Samples analysed	4: Technique	5: Column	6: Sample intake (g)
1001	a) Yes	a) Yes	B) 10-50	E) HPLC-FD	Vydac 201 TP54 4.6 x 250 mm	2
1002	a) Yes	b) No	A) <10	E) HPLC-FD	Agilent Zorbax Eclipse Plus C18 3.5µm 100x4.6mm	5
1004	b) No	b) No	A) <10	F) HPLC-UV/FD	Grace Vydac, C18 250x4.6 mm, 5um	4
1005	a) Yes	b) No	A) <10	E) HPLC-FD	Waters PAH C18 5um, 3x250mm	5
1007						
1008	b) No	b) No	A) <10	G) LC-MS	Zorbax Eclipse PAH 2.1x50mm 1.8µ	5
1009	a) Yes	b) No	A) <10	B) GC-MS	ZB-35, ZB-5MS	2
1010	a) Yes	a) Yes	D) >100	E) HPLC-FD	PAH C18, ODS, 4.6 mm x 250 mm, 5 µm particle size	5
1011	a) Yes	b) No	A) <10	C) GC-MS/MS	Agilent Select PAH	0.5
1012	a) Yes	a) Yes	B) 10-50	C) GC-MS/MS	PAH Select column	1
1013	a) Yes	a) Yes	B) 10-50	F) HPLC-UV/FD	VYDAC 201TP54 250mm ID 4.6mm 5u	5
1014	a) Yes	a) Yes	A) <10	E) HPLC-FD	RESTEK Pinnacle IIPAH 4µm 150*4,6 mm	5
1015	a) Yes	b) No	A) <10	C) GC-MS/MS	Select PAH 15 x 0.15x(0.10)	2
1016	a) Yes	b) No	B) 10-50	B) GC-MS	60m x 0.25 x 0.25 DB5MS	10
1017	a) Yes	b) No	A) <10	B) GC-MS	DB 17 30m x 0.25mm x 0.15 um (50% Phenyl) - methylpolysilossane	1
1018	b) No	b) No	A) <10	E) HPLC-FD	LiChroCART 250-4 LiChrospher PAH (5 µm)	15
1020	a) Yes	b) No	A) <10	B) GC-MS	Zebron Capiillary GC Column 30m×0.25mm×0.25µm	10
1022	a) Yes	b) No	A) <10	D) GC-HRMS	PAH select (varian)	2
1023	b) No	b) No	A) <10	C) GC-MS/MS	CP7461 varian Select PAH, 15x15	0.5
1024	a) Yes	a) Yes	A) <10	E) HPLC-FD	Agilent (Varian) PAH Pursuit 250 x 4.6 mm, 5 um	5
1025	b) No	b) No	A) <10	E) HPLC-FD	Waters PAH C 18, S-5µm; 250 x 3.0 µm	5
1027	a) Yes	a) Yes	B) 10-50	B) GC-MS	DB 35ms, 30m, i.d. 0.25mm, 0.15µm	5
1028	a) Yes	b) No	A) <10	B) GC-MS	Select PAH (30mx0.25mmx0.15um)	2
1029	a) Yes	a) Yes	A) <10	E) HPLC-FD	PAH C18 5um; 4,6x250mm, 5 µm (Waters P/N 186001265)	1
1030	a) Yes	a) Yes	B) 10-50	B) GC-MS	Varian Select	5

continued:

Participant	1: Accreditation	2: within Scope	3:Samples analysed	4: Technique	5: Column	6: Sample intake (g)
6001	b) No	b) No	A) <10	E) HPLC-FD	Restek Pinnacle II PAH	2
6002	b) No	b) No	A) <10	B) GC-MS	DB-EUPAH (Agilent)	5
6003	a) Yes	b) No	A) <10	C) GC-MS/MS	Varian select PAH 30m*0.25 mm 0.25 µm	5
6004	a) Yes	a) Yes	B) 10-50	C) GC-MS/MS	ZEBRON ZB 5MS 30 M 0.25/0.25/	1
6005	a) Yes	a) Yes	A) <10	C) GC-MS/MS	VF 17 MS	1
6006	b) No	b) No	A) <10	C) GC-MS/MS	AGILENT SELECT PAH (30mx0.25mmx0.15µm)	1
6007						
6008	a) Yes	b) No	A) <10	C) GC-MS/MS	SELECT PAH	5
6010	a) Yes	a) Yes	A) <10	E) HPLC-FD	VYDAC	10
6011	a) Yes	a) Yes	A) <10	D) GC-HRMS	VF-17ms (60 m x 0.25 mm x 0.25 µm)	5
6012	a) Yes	a) Yes	B) 10-50	B) GC-MS	Select PAH 15 x 0.15 (0.10) ; Agilent CP7461	2
6013	a) Yes	a) Yes	B) 10-50	E) HPLC-FD	Supelcosil PAH	9
6014	a) Yes	a) Yes	B) 10-50	C) GC-MS/MS	Agilent Select PAH; 30 m x 0.24 mm ID x 0.15 µm	2
6015	a) Yes	b) No	B) 10-50	E) HPLC-FD	Nucleosil 100-5 C18 PAH	5
6017	a) Yes	a) Yes	A) <10	F) HPLC-UV/FD	Pursuit 5 PAH, 250 x 4,6	1
6018	a) Yes	a) Yes	A) <10	I) UHPLC-FD	Nucleodur-PAH	1
6019	a) Yes	a) Yes	B) 10-50	C) GC-MS/MS	VF-17MS 15m ID 0.75mm Film 0.15µm	5
6020	b) No	b) No	B) 10-50	F) HPLC-UV/FD	Nucleosil 100-5 C18 PAH 250x3 mm	5
6021	b) No	b) No	A) <10	E) HPLC-FD	Grace VyDAC Typ 201P54	5
6022	b) No	b) No	A) <10	E) HPLC-FD	Pinnacle II PAH 4um 150x3.2mm	10
6024	b) No	b) No	A) <10	B) GC-MS	HP5 MSI	5
6026	b) No	b) No	A) <10	B) GC-MS	ZB-50 (Phenomenex)	10

7) Which of the following sample preparation procedures did you apply for the chocolate sample?
7.1) If applicable: Which extraction technique was applied?
7.2) If applicable: Which chromatography/fractionation technique was applied?
7.2.1) If applicable: Please specify SPE cartridge(s)

Participant	7: Sample preparation principle	7.1: Extraction technique	7.2: Chromatography / fractionation technique	7.2.1: SPE cartridges
1001	1) Extraction with organic solvent	A) Pressurised liquid extraction (PLE)	B) Gel permeation chromatography (GPC)	
1002	1) Extraction with organic solvent, 2) Liquid/Liquid partitioning, 4) Chromatography/fractionation	D) Other	C) Donor acceptor complex chromatography (DACC)	
1004	3) Saponification	X	X	C18 2g 12 mL, Florisil 500mg 3mL
1005	1) Extraction with organic solvent, 4) Chromatography/fractionation	C) Soxhlet extraction	B) Gel permeation chromatography (GPC)	
1007				
1008	1) Extraction with organic solvent	A) Pressurised liquid extraction (PLE)	B) Gel permeation chromatography (GPC)	
1009	1) Extraction with organic solvent, 4) Chromatography/fractionation	B) Sonication	B) Gel permeation chromatography (GPC)	
1010	1) Extraction with organic solvent	X	B) Gel permeation chromatography (GPC)	
1011	1) Extraction with organic solvent	A) Pressurised liquid extraction (PLE)	D) Solid phase extraction (SPE)	Supelco Envi-ChromP
1012	1) Extraction with organic solvent, 4) Chromatography/fractionation	A) Pressurised liquid extraction (PLE)	D) Solid phase extraction (SPE)	Envi Chrom-P
1013	3) Saponification	X	D) Solid phase extraction (SPE)	c18 2g 12 ml , Florizil 500mg 3 ml
1014	1) Extraction with organic solvent	B) Sonication	D) Solid phase extraction (SPE)	
1015	2) Liquid/Liquid partitioning, 4) Chromatography/fractionation	X	A) Column chromatography on silica	
1016	2) Liquid/Liquid partitioning, 3) Saponification	D) Other	A) Column chromatography on silica	
1017	1) Extraction with organic solvent	D) Other	D) Solid phase extraction (SPE)	SupelMIP 50mg/3ml
1018	2) Liquid/Liquid partitioning, 3) Saponification, 4) Chromatography/fractionation	X	A) Column chromatography on silica	
1020	3) Saponification	D) Other	D) Solid phase extraction (SPE)	Strata SI-1 Silica (55µm, 70A) 500 mg/6 mL
1022	1) Extraction with organic solvent	A) Pressurised liquid extraction (PLE)	B) Gel permeation chromatography (GPC), D) Solid phase extraction (SPE)	aluminiumoxide (14% H2O)
1023	1) Extraction with organic solvent, 4) Chromatography/fractionation	A) Pressurised liquid extraction (PLE)	D) Solid phase extraction (SPE)	ENVI Chrom P SPE tubes
1024	1) Extraction with organic solvent, 3) Saponification, 4) Chromatography/fractionation	D) Other	D) Solid phase extraction (SPE)	1st C18 2 g / 6 ml; 2nd silicagel 1 g / 6 ml
1025	2) Liquid/Liquid partitioning, 3) Saponification	X	X	
1027	1) Extraction with organic solvent, 3) Saponification	D) Other	D) Solid phase extraction (SPE)	Silica 5g (Strata) and PAH-HC 1g (Isolute)
1028	1) Extraction with organic solvent	B) Sonication	B) Gel permeation chromatography (GPC), D) Solid phase extraction (SPE)	PSA cartridge
1029	1) Extraction with organic solvent	A) Pressurised liquid extraction (PLE)	B) Gel permeation chromatography (GPC)	
1030	1) Extraction with organic solvent, 2) Liquid/Liquid partitioning, 3) Saponification, 4) Chromatography/fractionation	X	A) Column chromatography on silica	

continued:

Participant	7: Sample preparation principle	7.1: Extraction technique	7.2: Chromatography / fractionation technique	7.2.1: SPE cartridges
6001	3) Saponification	X	X	Bakerbond SPE Silica / Bakerbond SPE Cyano
6002	1) Extraction with organic solvent	D) Other	X	
6003	1) Extraction with organic solvent	C) Soxhlet extraction	D) Solid phase extraction (SPE)	supelco Envi chrom P
6004	1) Extraction with organic solvent	X	D) Solid phase extraction (SPE)	
6005	1) Extraction with organic solvent	A) Pressurised liquid extraction (PLE)	A) Column chromatography on silica	
6006	1) Extraction with organic solvent	D) Other	D) Solid phase extraction (SPE)	envichromP
6007				
6008	1) Extraction with organic solvent, 4) Chromatography/fractionation	A) Pressurised liquid extraction (PLE)	D) Solid phase extraction (SPE)	ENVI CHROM P SUPELCO-57226 0.5g 6 ml
6010	1) Extraction with organic solvent, 2) Liquid/Liquid partitioning, 3) Saponification	D) Other	X	
6011	2) Liquid/Liquid partitioning, 3) Saponification, 4) Chromatography/fractionation	X	E) Other	
6012	2) Liquid/Liquid partitioning	B) Sonication	D) Solid phase extraction (SPE)	
6013	3) Saponification	D) Other	D) Solid phase extraction (SPE)	Silica; 55 µm; 70A
6014	1) Extraction with organic solvent, 2) Liquid/Liquid partitioning, 3) Saponification	B) Sonication	A) Column chromatography on silica	
6015	1) Extraction with organic solvent	C) Soxhlet extraction	D) Solid phase extraction (SPE)	Chromabond HR-P 6ml/500mg
6017	1) Extraction with organic solvent, 3) Saponification, 4) Chromatography/fractionation	B) Sonication	B) Gel permeation chromatography (GPC)	
6018	1) Extraction with organic solvent, 2) Liquid/Liquid partitioning, 3) Saponification	B) Sonication	X	
6019	1) Extraction with organic solvent, 4) Chromatography/fractionation	A) Pressurised liquid extraction (PLE)	B) Gel permeation chromatography (GPC), D) Solid phase extraction (SPE), E) Other	Florisil
6020	2) Liquid/Liquid partitioning, 3) Saponification, 4) Chromatography/fractionation	X	A) Column chromatography on silica	
6021	2) Liquid/Liquid partitioning, 3) Saponification, 4) Chromatography/fractionation	X	D) Solid phase extraction (SPE)	Strata SDB-L Styrene-Divenylbenzene, Phenomenex
6022	2) Liquid/Liquid partitioning, 3) Saponification, 4) Chromatography/fractionation	D) Other	D) Solid phase extraction (SPE)	Discovery SPE DSC-SI Silica Tube 12ml, 2g
6024	1) Extraction with organic solvent	X	D) Solid phase extraction (SPE)	Bond Elut C18 and Florisil
6026	1) Extraction with organic solvent, 3) Saponification, 4) Chromatography/fractionation	D) Other	A) Column chromatography on silica	deuterated

8) How did you calibrate your instrument?
8.1) Did you apply external calibration?
8.1.1) In case of external calibration: How did you calibrate?
8.2) Did you apply internal standardization?
8.2.1) Which internal standards did you apply?
8.2.2) Please provide details on internal standards
8.3) Did you apply standard addition?

Participant	8.1: External calibration	8.1.1: Details on external calibration	8.2: Internal standardisation	8.2.1: Type of internal standards	8.2.2: Details on internal standards	8.3: Standard addition
1001	a) Yes	A) with standards in an organic solvent	b) No	X		b) No
1002	b) No	X	a) Yes	B) Stable isotope labelled analogue(s)	DiP D14	a) Yes
1004	a) Yes	A) with standards in an organic solvent	b) No	X		b) No
1005	a) Yes	A) with standards in an organic solvent	b) No	X		a) Yes
1007						
1008	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	carbon 13 labelled standards from ehrensdorfer	b) No
1009	a) Yes	B) with matrix matched standards	b) No	X		a) Yes
1010	a) Yes	A) with standards in an organic solvent	b) No	X		a) Yes
1011	b) No	X	a) Yes	B) Stable isotope labelled analogue(s)	13C	a) Yes
1012	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	Bap 13C4, CHR 13C6, BbF 13C6, BaA 13C6	b) No
1013	b) No	X	a) Yes	A) Structural analogue(s) of the analyte(s)	Benzo (b) chrysene	a) Yes
1014	a) Yes	A) with standards in an organic solvent	b) No	X		a) Yes
1015	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	Benzo(a)pyrene-D12, Chrysene-D12	b) No
1016	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	Ehrenstorfer deuterated mix 9 + D14 DaiP & 13C6DaeP	b) No
1017	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	BaA d12; CHR d12; BbF d12; BaP d12	a) Yes
1018	b) No	X	a) Yes	A) Structural analogue(s) of the analyte(s)	Benzo(b)Chrysene	b) No
1020	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	Benzo(a)pyrene-D12; Benzo(b)fluoranthene-D12; Chrysene-D12; Benzo(a)anthracene-D12	a) Yes
1022	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	BAA-D12, CHR-D12, BBF-D12, BAP-D12	b) No
1023	a) Yes	B) with matrix matched standards	a) Yes	B) Stable isotope labelled analogue(s)	13C-marked	b) No
1024	a) Yes	A) with standards in an organic solvent	a) Yes	A) Structural analogue(s) of the analyte(s)	benzo(b)chrysene, but only for assessment of extraction and saponification step yield, not for calculation of PAHs amounts	b) No
1025	a) Yes	A) with standards in an organic solvent	b) No	X		b) No
1027	b) No	X	a) Yes	B) Stable isotope labelled analogue(s)	Mix of 9 deuterated PAH standards	b) No
1028	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	B(a)A-D12, B(a)P-D12, CHR-D12, B(b)F-D12	a) Yes
1029	a) Yes	A) with standards in an organic solvent	b) No	X		a) Yes
1030	b) No	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	US EPA16 13C PAH Cocktail	a) Yes

continued:

Participant	8.1: External calibration	8.1.1: Details on external calibration	8.2: Internal standardisation	8.2.1: Type of internal standards	8.2.2: Details on internal standards	8.3: Standard addition
6001	a) Yes	A) with standards in an organic solvent	b) No	X		b) No
6002	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	Benz[a]pyren D12	a) Yes
6003	b) No	X	a) Yes	B) Stable isotope labelled analogue(s)	the internal standards are added to the beginning of sample handling	b) No
6004	b) No	X	a) Yes	B) Stable isotope labelled analogue(s)	1	b) No
6005	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	4	b) No
6006	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	calcul excel	b) No
6007						
6008	b) No	X	a) Yes	B) Stable isotope labelled analogue(s)	EACH ISOTOPE OF ANALYTE	a) Yes
6010	a) Yes	A) with standards in an organic solvent	a) Yes	A) Structural analogue(s) of the analyte(s)	6-methylchrysene	a) Yes
6011	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	d12-benzo(a)anthracene, d12-chrysene, d12-benzo(b)fluoranthene, d12-benzo(a)pyrene	b) No
6012	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	d12-Chrysene for BAA and CHR; d12-Benzo(a)pyrene for BFB and BAP	b) No
6013	a) Yes	A) with standards in an organic solvent	a) Yes	A) Structural analogue(s) of the analyte(s)	6-Methyl-Chrysene	a) Yes
6014	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	13 C labelled PAH4	b) No
6015	a) Yes	A) with standards in an organic solvent	a) Yes	A) Structural analogue(s) of the analyte(s)	Benzo(b)chrysen	b) No
6017	a) Yes	A) with standards in an organic solvent	a) Yes	A) Structural analogue(s) of the analyte(s)	Benzo(b)chrysen	b) No
6018	a) Yes	A) with standards in an organic solvent	a) Yes	A) Structural analogue(s) of the analyte(s)	Benzo(b)chrysene	b) No
6019	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	13C-labelled analogues	b) No
6020	a) Yes	A) with standards in an organic solvent	a) Yes	A) Structural analogue(s) of the analyte(s)	Benzo(b)chrysen	b) No
6021	a) Yes	A) with standards in an organic solvent	b) No	X		a) Yes
6022	a) Yes	A) with standards in an organic solvent	b) No	X		b) No
6024	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	d10 phenanthrene	b) No
6026	a) Yes	A) with standards in an organic solvent	a) Yes	B) Stable isotope labelled analogue(s)	deuterated	b) No

9.) How did you obtain the PAH contents of the chocolate sample expressed on fat basis?
10) How did you determine the fat content of the chocolate sample?

Participant	9:. How were analyte contents expressed on fat basis obtained?	10: Fat content determination
1001	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	D) Other
1002	B) Extracting fat from the chocolate matrix and performing analysis on the fat fraction	B) Only solvent extraction
1004	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1005	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1007		
1008	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	B) Only solvent extraction
1009	B) Extracting fat from the chocolate matrix and performing analysis on the fat fraction	B) Only solvent extraction
1010	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1011	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1012	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	B) Only solvent extraction
1013	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1014	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	B) Only solvent extraction
1015	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1016	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1017	B) Extracting fat from the chocolate matrix and performing analysis on the fat fraction	A) Hydrolysis of the sample followed by solvent extraction
1018	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1020	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1022	B) Extracting fat from the chocolate matrix and performing analysis on the fat fraction	B) Only solvent extraction
1023	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	B) Only solvent extraction
1024	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1025	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1027	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1028	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1029	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
1030	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	D) Other

continued:

Participant	9:. How were analyte contents expressed on fat basis obtained?	10: Fat content determination
6001	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	D) Other
6002	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
6003	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
6004	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	B) Only solvent extraction
6005		A) Hydrolysis of the sample followed by solvent extraction
6006	B) Extracting fat from the chocolate matrix and performing analysis on the fat fraction	B) Only solvent extraction
6007		
6008	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	B) Only solvent extraction
6010	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
6011	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	D) Other
6012	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	B) Only solvent extraction
6013	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
6014	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	B) Only solvent extraction
6015	B) Extracting fat from the chocolate matrix and performing analysis on the fat fraction	B) Only solvent extraction
6017	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
6018	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
6019	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	B) Only solvent extraction
6020	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
6021	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	A) Hydrolysis of the sample followed by solvent extraction
6022	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	B) Only solvent extraction
6024	A) Correcting of the PAH content of the chocolate sample (expressed in µg/kg chocolate) with a separately determined ratio for the fat content.	B) Only solvent extraction
6026	B) Extracting fat from the chocolate matrix and performing analysis on the fat fraction	B) Only solvent extraction

11) Did you experience any problems during sample preparation of the chocolate/cocoa butter sample?
11.1) Please specify:
12) Did you experience any chromatographic interferences?
12.1) Please specify:

Participant	11: Problems during sample preparation	11.1: Details on experienced problems	12: Chromatographic interferences	12.1: Details on chromatographic interferences
1001	b) No		b) No	
1002	b) No		a) Yes	We validated our method with chocolate previously and we had any interference. However in these samples, we had an interference for B[a]A. So for this PAH, we applied the standard addition.
1004	b) No		b) No	
1005	b) No		a) Yes	matrix interferences mainly with BAA
1007				
1008	a) Yes	one sample resulted in problems on GPC, technical problem not sample related.	a) Yes	benzo[a]pyrene internal standard resulted in problems on detection in samples
1009	b) No		b) No	
1010	b) No		b) No	
1011	b) No		b) No	
1012	b) No		b) No	
1013	b) No		a) Yes	in chrysene and in benzo(a)anthracene
1014	b) No		b) No	
1015	b) No		b) No	
1016	b) No		b) No	
1017	b) No		b) No	
1018	b) No		b) No	
1020	a) Yes	After saponification, chocolate quickly became hard. So, it was impossible to filter the sample throught the glass filter with pores.	b) No	
1022	b) No		b) No	
1023	b) No		b) No	
1024	b) No		b) No	
1025	b) No		b) No	
1027	a) Yes	The sample has to be kept warm during the preparation. If it cools to room temperature it becomes a gel.	b) No	
1028	b) No		b) No	
1029	b) No		a) Yes	chrysene
1030	b) No		b) No	

continued:

Participant	11: Problems during sample preparation	11.1: Details on experienced problems	12: Chromatographic interferences	12.1: Details on chromatographic interferences
6001	a) Yes	Bad recoveryfaktors with more than 2g of sample ammount.	a) Yes	Matrixinterferences at retentiontime from crysene.
6002	b) No		b) No	
6003	b) No		b) No	
6004	b) No		b) No	
6005			b) No	
6006	b) No		b) No	
6007				
6008	b) No		b) No	
6010	b) No		b) No	
6011	b) No		b) No	
6012	b) No		a) Yes	samples contains Triphenylene which elutes nearby Chrysene; Peakresolution about 50%
6013	b) No		b) No	
6014	b) No		b) No	
6015	a) Yes	solubility problem of cocoabutter fat with the solvent for SPE cleanup	a) Yes	coelutions with Chrysen and Benzo(a)anthracen
6017	a) Yes	It was not a chocolate. We've tried to use a sample amount of 5 g, but it was not our method	b) No	
6018	b) No		b) No	
6019	b) No		b) No	
6020	b) No		b) No	
6021	a) Yes	Chocolate: stable emulsions during liquid/liquid partitioning, difficulties in separating layers	b) No	
6022	a) Yes	Difficult to achieve clear separation between aqueous and organic layer during the liquid-liquid partitioning stage of the extraction procedure with the chocolate sample. This was not experienced when extracting the butter sample.	b) No	
6024	b) No		a) Yes	interfering peaks - possiblly inaqeduate sample clean up?
6026	b) No		b) No	

METHOD PERFORMANCE PARAMETERS

With reference to Commission Regulation (EC) No 333/2007 as amended by Commission Regulation (EU) No 836/2011, non-compliant method performance characteristics are marked in the tables in bold red font. Threshold values for the evaluation were LOD≤ 0.30 µg/kg, LOQ ≤ 0.90 µg/kg.
Despite it was requested to express recovery as a yield of the assay, many participants seemed to have reported apparent recovery values. Due to this inconsistency in reporting, recovery values were not rated.

Method performance data reported by participants for the determination of BAA and BAP in cocoa butter.

	BAA				BAP		
Participant	LOD	LOQ	Recovery	Participant	LOD	LOQ	Recovery
	µg/kg	µg/kg	%		µg/kg	µg/kg	%
1001	0.06	0.2	100	1001	0.06	0.2	100
1002	0.2	0.4	102.3	1002	0.2	0.4	100.2
1004	0.1	0.33	107.5	1004	0.1	0.33	86.1
1005	0.025	0.05	102	1005	0.025	0.05	97
1007	n.r.	n.r.	n.r.	1007	n.r.	n.r.	n.r.
1008	0.1	0.3	58.9	1008	0.1	0.3	83.3
1009	0.3	**1.1**	100	1009	0.2	0.6	94
1010	0.03	0.44	88.7	1010	0.01	0.44	86.6
1011	0.3	0.9	97	1011	0.3	0.9	94
1012	0.03	0.06	82	1012	0.02	0.05	75
1013	0.1	0.3	104	1013	0.1	0.3	105
1014	0.1	0.5	80	1014	0.04	0.2	80
1015	**0.45**	**1.5**	79.4	1015	0.15	0.5	99
1016	0.2	0.6	95.8	1016	0.2	0.7	98.5
1017	0.07	0.2	84	1017	0.07	0.2	92
1018	0.2	0.6	102	1018	0.1	0.2	88
1020	0.19	0.64	89	1020	0.21	0.71	96
1022	0.009	0.018	85	1022	0.004	0.008	85
1023	n.r.	n.r.	n.r.	1023	n.r.	n.r.	n.r.
1024	0.11	0.21	92.9	1024	0.09	0.18	94.4
1025	0.08	0.25	96	1025	0.08	0.25	71
1027	0.13	0.38	54	1027	0.14	0.41	53
1028	**0.5**	**1**	87	1028	**0.5**	**1**	83
1029	0.07	0.21	97	1029	0.08	0.24	94
1030	0.01	0.01	54	1030	0.08	0.08	57
6001	0.3	0.6	87	6001	0.3	0.6	93
6002	0.3	0.5	104	6002	0.3	0.5	94
6003	0.1	0.2	80	6003	0.1	0.2	86
6004	0.03	0.1	83.6	6004	0.03	0.1	72.9
6005	0.1	0.5	90	6005	0.1	0.5	95
6006	0.03	0.099	75	6006	0.03	0.099	69
6007	n.r.	n.r.	n.r.	6007	n.r.	n.r.	n.r.
6008	0.03	0.1	99	6008	0.03	0.1	92
6010	0.3	0.5	n.r.	6010	0.3	0.5	n.r.
6011	0.06	0.12	89	6011	0.15	0.31	95
6012	0.3	0.5	80	6012	0.3	0.5	80
6013	0.1	0.2	56.9	6013	0.1	0.2	56.9
6014	0.04	0.08	83	6014	0.04	0.08	100
6015	0.1	0.2	105	6015	0.1	0.2	100
6017	**0.5**	**1**	74	6017	**0.5**	**1**	109
6018	0.05	0.1	>50	6018	0.05	0.1	>50
6019	0.02	0.05	90	6019	0.02	0.05	86
6020	0.02	0.11	85.6	6020	0.03	0.15	86.6
6021	0.3	0.5	89	6021	0.3	0.5	86
6022	**0.5**	**5**	45	6022	**0.5**	**5**	35
6024	0.1	0.3	81.2	6024	0.1	0.3	81.2
6026	**0.5**	**1**	51	6026	**0.5**	0.9	51

n.r.: not reported

Method performance data reported by participants for the determination of BBF and CHR in cocoa butter.

	BBF				CHR		
Participant	LOD	LOQ	Recovery	Participant	LOD	LOQ	Recovery
	µg/kg	µg/kg	%		µg/kg	µg/kg	%
1001	0.1	0.3	98	1001	0.03	0.1	103
1002	0.2	0.4	99.1	1002	0.2	0.4	107
1004	0.1	0.33	95.4	1004	0.25	0.83	81.1
1005	0.05	0.1	86	1005	0.025	0.05	90
1007	n.r.	n.r.	n.r.	1007	n.r.	n.r.	n.r.
1008	0.1	0.3	68.8	1008	0.1	0.3	73.4
1009	0.3	0.9	104	1009	0.1	0.4	109
1010	0.035	0.45	93.4	1010	0.023	0.45	89.4
1011	0.3	0.9	96	1011	0.3	0.9	101
1012	0.02	0.08	78	1012	0.03	0.06	77
1013	0.1	0.3	105	1013	0.2	0.6	79
1014	0.04	0.2	80	1014	0.1	0.5	80
1015	0.28	**0.94**	91.6	1015	**0.36**	**1.2**	89.3
1016	0.2	0.5	95	1016	0.1	0.3	100
1017	0.07	0.2	103	1017	0.07	0.2	89
1018	0.2	0.6	88	1018	0.2	0.6	107
1020	0.16	0.52	94	1020	0.17	0.56	105
1022	0.003	0.006	83	1022	0.006	0.012	74
1023	n.r.	n.r.	n.r.	1023	n.r.	n.r.	n.r.
1024	0.21	0.41	96	1024	0.11	0.22	94.9
1025	0.09	0.26	83	1025	0.08	0.25	99
1027	0.23	0.68	53	1027	0.13	0.4	50
1028	**0.5**	**1**	85	1028	**0.5**	**1**	89
1029	0.11	0.33	95	1029	0.12	0.36	94
1030	0.06	0.06	55	1030	0.06	0.06	52
6001	0.3	0.6	88	6001	0.3	0.6	104
6002	0.3	0.5	94	6002	0.3	0.5	101
6003	0.1	0.2	106	6003	0.1	0.2	76
6004	0.03	0.1	91.7	6004	0.03	0.1	82.6
6005	0.1	0.5	105	6005	0.1	0.5	115
6006	0.03	0.099	78	6006	0.03	0.099	75
6007	n.r.	n.r.	n.r.	6007	n.r.	n.r.	n.r.
6008	0.03	0.1	98	6008	0.03	0.1	99
6010	0.3	0.5	n.r.	6010	0.3	0.5	n.r.
6011	0.06	0.13	93	6011	0.06	0.13	95
6012	0.3	0.5	80	6012	0.3	0.5	80
6013	0.1	0.2	56.9	6013	0.1	0.2	56.9
6014	0.1	0.2	95	6014	0.1	0.2	86
6015	0.2	0.4	125	6015	0.1	0.2	95
6017	**0.5**	**1**	101	6017	**0.5**	**1**	101
6018	0.2	0.4	>50	6018	0.2	0.4	>50
6019	0.02	0.05	84	6019	0.02	0.05	91
6020	0.06	0.27	88.1	6020	0.04	0.21	87.3
6021	0.3	0.5	86	6021	**0.5**	0.9	83
6022	**0.5**	**5**	23	6022	**0.5**	**5**	52
6024	0.1	0.3	81.2	6024	0.1	0.3	81.2
6026	**0.5**	**1**	51	6026	**0.5**	**1**	51

n.r.: not reported

Method performance data reported by participants for the determination of BAA and BAP in chocolate.

	BAA				BAP		
Participant	LOD	LOQ	Recovery	Participant	LOD	LOQ	Recovery
	µg/kg	µg/kg	%		µg/kg	µg/kg	%
1001	0.06	0.2	92	1001	0.06	0.2	92
1002	0.2	0.4	91.2	1002	0.2	0.4	85.7
1004	0.1	0.33	86.8	1004	0.1	0.33	71
1005	0.025	0.05	96	1005	0.025	0.05	97
1007	n.r.	n.r.	n.r.	1007	n.r.	n.r.	n.r.
1008	0.25	0.75	58.9	1008	0.25	0.75	83.3
1009	0.3	1.1	100	1009	0.2	0.6	94
1010	0.01	0.4	104.2	1010	0.003	0.4	98.3
1011	0.3	0.9	95	1011	0.3	0.9	102
1012	0.03	0.06	63	1012	0.02	0.05	74
1013	0.1	0.3	110	1013	0.1	0.3	103
1014	0.1	0.5	80	1014	0.04	0.2	80
1015	0.38	1.28	74.5	1015	0.17	0.57	87.3
1016	0.2	0.3	95.8	1016	0.3	0.5	98.5
1017	0.06	0.2	78	1017	0.06	0.2	85
1018	0.2	0.6	104	1018	0.1	0.4	96
1020	0.25	0.83	110	1020	0.22	0.74	92
1022	0.006	0.012	88	1022	0.004	0.008	81
1023	n.r.	n.r.	n.r.	1023	n.r.	n.r.	n.r.
1024	0.11	0.21	91.3	1024	0.09	0.18	89.8
1025	0.03	0.1	87	1025	0.03	0.1	77
1027	0.13	0.38	54	1027	0.14	0.41	53
1028	0.5	1.25	106	1028	0.5	1.25	106
1029	0.08	0.24	95	1029	0.07	0.21	96
1030	0.01	0.01	75	1030	0.08	0.08	71
6001	0.3	0.6	41	6001	0.3	0.6	55
6002	0.3	0.5	94	6002	0.3	0.5	93
6003	0.02	0.05	70	6003	0.02	0.05	70
6004	0.03	0.1	71.8	6004	0.03	0.1	73.7
6005	0.1	0.5	105	6005	0.1	0.5	95
6006	0.029	0.098	76	6006	0.029	0.098	68
6008	0.04	0.11	95	6008	0.04	0.11	94
6010	0.3	0.5	n.r.	6010	0.3	0.5	n.r.
6011	0.06	0.12	89	6011	0.15	0.31	95
6012	0.3	0.5	80	6012	0.3	0.5	80
6013	0.1	0.2	52.8	6013	0.1	0.2	54.9
6014	0.02	0.05	83	6014	0.02	0.05	86
6015	0.2	0.4	105	6015	0.2	0.4	100
6017	0.5	1	96	6017	0.5	1	120
6018	0.05	0.1	>50	6018	0.05	0.1	>50
6019	0.02	0.05	100	6019	0.02	0.05	92
6020	0.02	0.11	85.6	6020	0.03	0.15	86.6
6021	0.3	0.5	92	6021	0.1	0.3	89
6022	0.5	5	18	6022	0.5	5	12
6024	0.1	0.3	74.6	6024	0.1	0.3	74.6
6026	0.5	1	68	6026	0.5	0.9	66

n.r.: not reported

Method performance data reported by participants for the determination of BBF and CHR in chocolate.

	BBF				CHR		
Participant	LOD	LOQ	Recovery	Participant	LOD	LOQ	Recovery
	µg/kg	µg/kg	%		µg/kg	µg/kg	%
1001	0.1	0.3	96	1001	0.03	0.1	97
1002	0.2	0.4	89	1002	0.2	0.4	102.2
1004	0.1	0.33	83.6	1004	0.25	0.83	77.6
1005	0.05	0.1	94	1005	0.025	0.05	93
1007	n.r.	n.r.	n.r.	1007	n.r.	n.r.	n.r.
1008	0.25	0.75	68.8	1008	0.25	0.75	73.4
1009	0.3	0.9	104	1009	0.1	0.4	109
1010	0.012	0.4	103.7	1010	0.008	0.4	99.8
1011	0.3	0.9	98	1011	0.3	0.9	93
1012	0.02	0.08	67	1012	0.03	0.06	65
1013	0.1	0.3	95	1013	0.2	0.6	101
1014	0.04	0.2	80	1014	0.1	0.5	80
1015	0.14	0.45	95.3	1015	0.19	0.63	77.4
1016	0.2	0.3	95	1016	0.1	0.2	100
1017	0.06	0.2	94	1017	0.06	0.2	80
1018	0.2	0.5	93	1018	0.2	0.7	120
1020	0.16	0.53	104	1020	0.25	0.83	91
1022	0.003	0.006	79	1022	0.006	0.012	76
1023	n.r.	n.r.	n.r.	1023	n.r.	n.r.	n.r.
1024	0.21	0.41	81.9	1024	0.11	0.22	90.4
1025	0.03	0.1	72	1025	0.03	0.1	87
1027	0.23	0.68	53	1027	0.13	0.4	50
1028	**0.5**	**1.25**	104	1028	**0.5**	**1.25**	103
1029	0.12	0.36	92	1029	0.1	0.3	97
1030	0.06	0.06	77	1030	0.06	0.06	68
6001	0.3	0.6	59	6001	0.3	0.6	69
6002	0.3	0.5	92	6002	0.3	0.5	94
6003	0.02	0.05	85	6003	0.02	0.05	70
6004	0.03	0.1	89.4	6004	0.03	0.1	75.6
6005	0.1	0.5	105	6005	0.1	0.5	110
6006	0.029	0.098	77	6006	0.029	0.098	77
6008	0.04	0.11	97	6008	0.04	0.11	93
6010	0.3	0.5		6010	0.3	0.5	n.r.
6011	0.06	0.13	93	6011	0.06	0.13	95
6012	0.3	0.5	80	6012	0.3	0.5	80
6013	0.1	0.2	50.5	6013	0.1	0.2	52.4
6014	0.05	0.1	80	6014	0.05	0.1	91
6015	**0.4**	0.8	125	6015	0.2	0.4	95
6017	**0.5**	**1**	115	6017	**0.5**	**1**	112
6018	0.2	0.4	>50	6018	0.2	0.4	>50
6019	0.02	0.05	92	6019	0.02	0.05	100
6020	0.06	0.27	88.1	6020	0.04	0.21	87.3
6021	0.3	0.5	87	6021	**0.4**	0.7	99
6022	**0.5**	**5**	10	6022	**0.5**	**5**	18
6024	0.1	0.3	74.6	6024	0.1	0.3	74.6
6026	**0.5**	**1**	66	6026	**0.5**	**1**	68

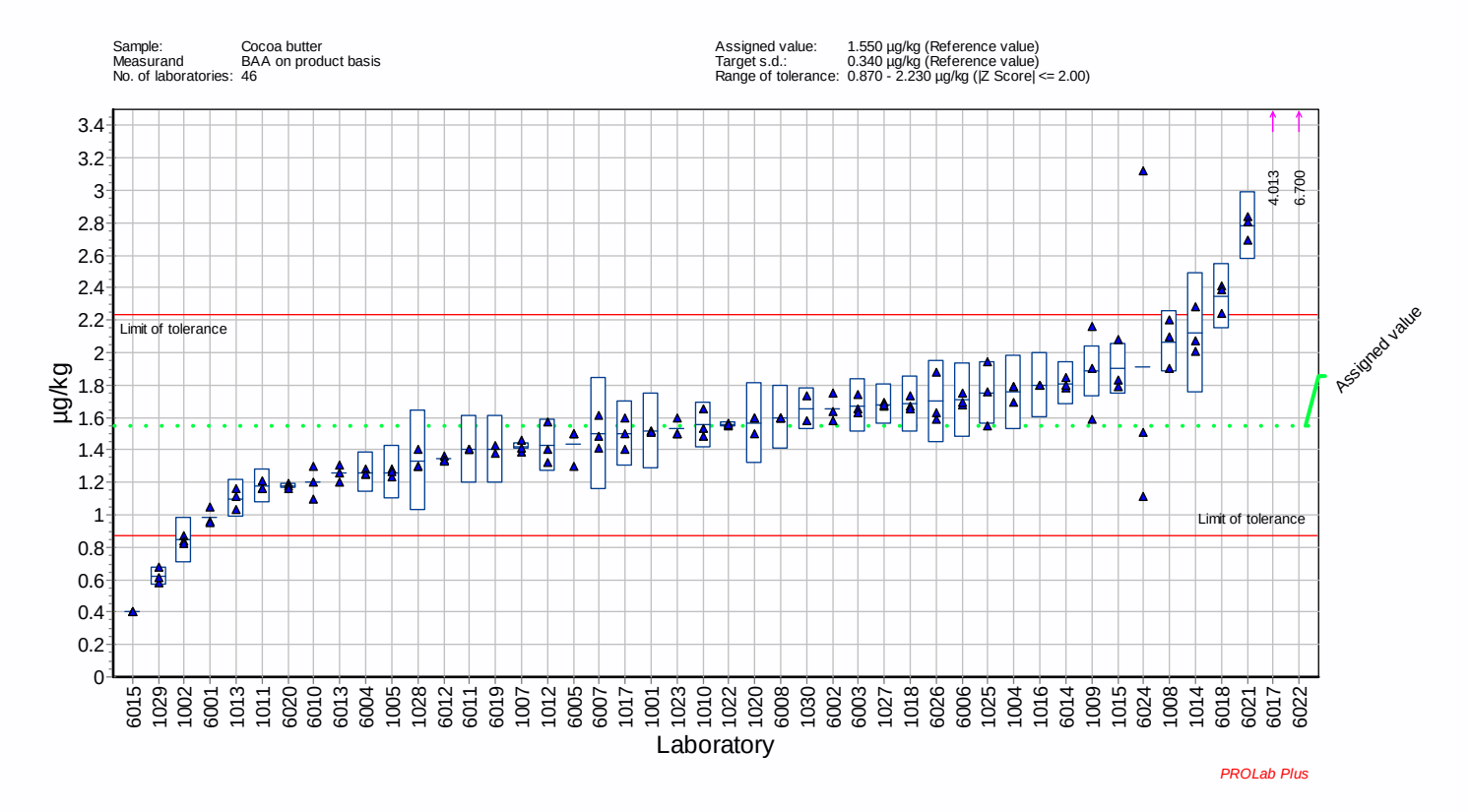
n.r.: not reported

ANNEX 8: Data reported by participants

The data reported by the participants are compiled in the following tables. Uncertainty values that do not comply with the U_f thresholds (individual PAHs), respectively that are not equal to the propagated uncertainties of the individual analytes (SUM4PAH parameter) are marked by bold red font. The results of replicate analyses together with the expanded measurement uncertainty (k=2) reported for the value for proficiency assessment are depicted in the graphs. Red lines indicate the thresholds for satisfactory z-scores.

Distribution of individual results of replicate determinations reported for the benz[*a*]anthracene (BAA) content of the cocoa butter test sample

blue triangles: individual results of replicate determinations, blue box: reported expanded measurement uncertainty (k=2), blue horizontal line in blue box: average of replicate determinations, green dotted line: assigned value, green area around assigned value: expanded uncertainty of the assigned value (k=2), red lines: lower and upper limit of satisfactory z-score range



Results reported by *NRLs* for the content of benz[*a*]anthracene (BAA) in the cocoa butter test material. Assigned value is 1.55 µg/kg

The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
1001	1.52	1.52	1.51	1.5	0.46	2
1002	0.871	0.840	0.819	0.843	0.283	2
1004	1.69	1.79	1.79	1.76	0.46	2
1005	1.28	1.23	1.27	1.26	0.33	2
1007	1.39	1.46	1.41	1.42	0.04	2
1008	2.2	1.9	2.1	2.2	0.4	2
1009	2.16	1.9	1.59	1.88	0.32	2
1010	1.65	1.53	1.48	1.56	0.28	2
1011	1.16	1.21	1.16	1.18	0.21	2
1012	1.40	1.57	1.32	1.43	0.32	2
1013	1.03	1.16	1.11	1.10	0.23	2
1014	2.28	2.01	2.07	2.12	0.74	2
1015	1.83	1.79	2.08	1.9	0.32	2
1016	1.8	1.8	1.8	1.8	0.4	2
1017	1.5	1.6	1.4	1.5	0.4	2
1018	1.67	1.73	1.65	1.68	0.35	2
1020	1.50	1.60	1.60	1.57	0.50	2
1022	1.551	1.567	1.555	1.557	0.027	2
1023	1.5	1.5	1.6	1.5	30	n.r.
1024	n.r.	n.r.	n.r.	n.r.	n.r.	n.r.
1025	1.76	1.94	1.55	1.75	0.39	2
1027	1.69	1.67	1.68	1.68	0.25	2
1028	1.3	1.3	1.4	1.3	0.6	2
1029	0.68	0.58	0.61	0.62	0.11	2
1030	1.58	1.73	n.r.	1.58	0.25	2

n.r.: not reported
k.: coverage factor
Rep.: result of replicate analysis

Results reported by *OCLs* for the content of benz[*a*]anthracene (BAA) in the cocoa butter test material. Assigned value is 1.55 µg/kg

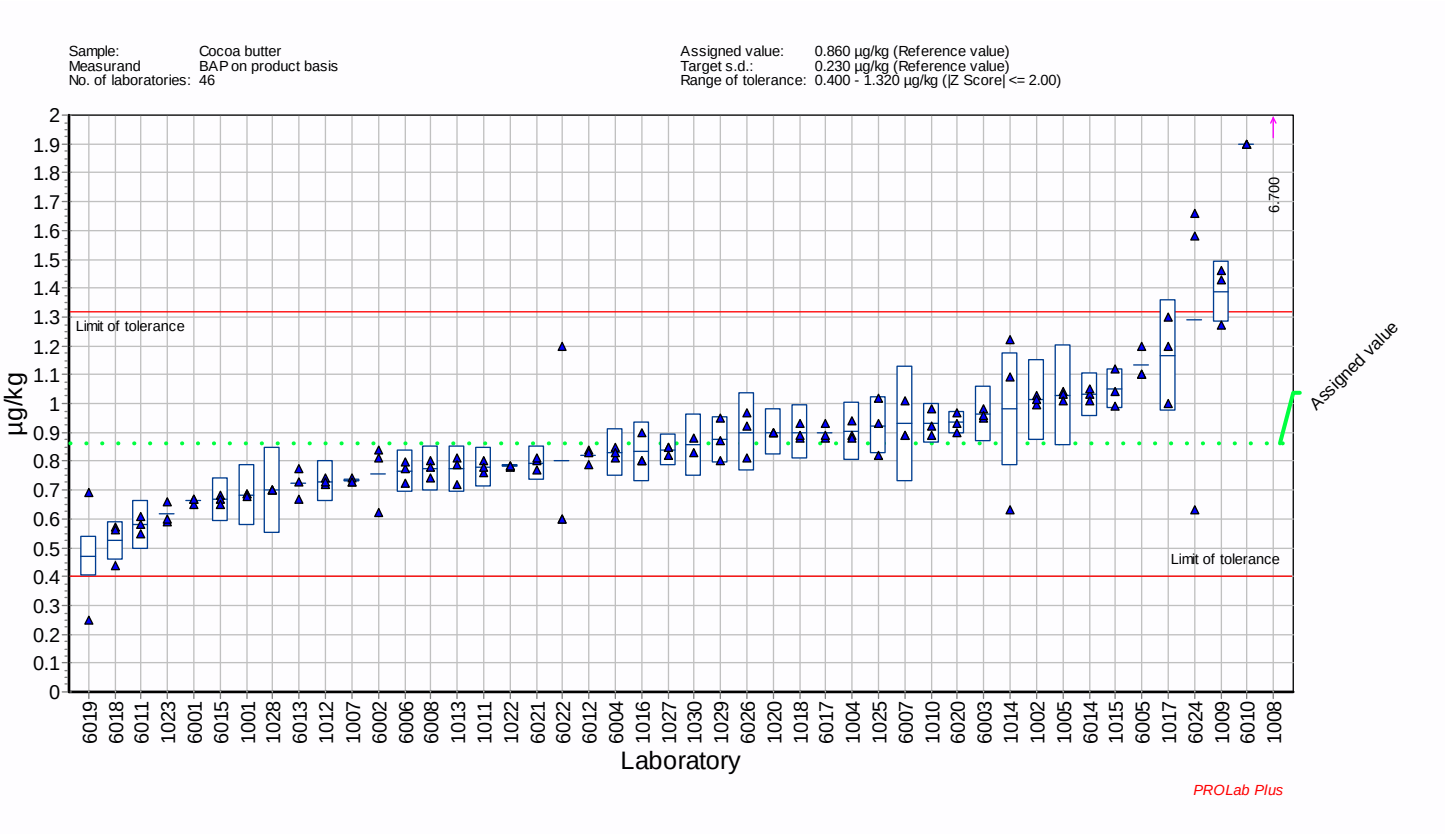
The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
6001	1.05	0.96	0.95	0.99	0.50	n.r.
6002	1.64	1.75	1.58	1.64	n.r.	n.r.
6003	1.65	1.63	1.74	1.67	0.33	2
6004	1.28	1.25	1.25	1.26	0.25	2
6005	1.3	1.5	1.5	1.4	25	2
6006	1.749	1.677	1.692	1.706	0.461	2
6007	1.41	1.48	1.61	1.50	0.70	2
6008	1.6	1.6	1.6	1.6	0.4	2
6010	1.2	1.3	1.1	1.2	n.r.	n.r.
6011	1.4	1.4	1.4	1.4	0.42	2
6012	1.36	1.34	1.33	1.34	20	n.r.
6013	1.20	1.31	1.26	1.26	35	56.9
6014	1.78	1.80	1.85	1.81	0.27	2
6015	<0.4	<0.4	<0.4	<0.4	n.r.	n.r.
6017	4.05	4.03	3.96	4.01	0.80	n.r.
6018	2.41	2.39	2.24	2.35	0.6	3
6019	1.43	1.38	n.r.	1.40	0.42	2
6020	1.19	1.18	1.16	1.18	0.03	2
6021	2.84	2.81	2.69	2.78	0.42	2
6022	6.3	7.0	6.8	6.7	0.21	n.r.
6024	1.11	1.51	3.12	1.91	n.r.	n.r.
6026	1.63	1.59	1.88	1.63	0.49	2

n.r.: not reported
k.: coverage factor
Rep.: result of replicate analysis

Distribution of individual results of replicate determinations reported for the benzo[*a*]pyrene (BAP) content of the cocoa butter test sample

blue triangles: individual results of replicate determinations, blue box: reported expanded measurement uncertainty (k=2), blue horizontal line in blue box: average of replicate determinations, green dotted line: assigned value, green area around assigned value: expanded uncertainty of the assigned value (k=2), red lines: lower and upper limit of satisfactory z-score range



Results reported by *NRLs* for the content of benzo[*a*]pyrene (BAP) in the cocoa butter test material. Assigned value is 0.86 µg/kg

The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
1001	0.687	0.687	0.676	0.68	0.21	2
1002	1.028	0.996	1.012	1.012	0.281	2
1004	0.94	0.89	0.88	0.90	0.20	2
1005	1.04	1.03	1.01	1.03	0.35	2
1007	0.73	0.74	0.73	0.73	0.01	2
1008	8.6	4.1	7.4	8.0	1.6	2
1009	1.27	1.43	1.46	1.39	0.21	2
1010	0.98	0.92	0.89	0.94	0.14	2
1011	0.80	0.76	0.78	0.78	0.14	2
1012	0.74	0.72	0.73	0.73	0.14	2
1013	0.72	0.81	0.79	0.77	0.16	2
1014	1.09	1.22	0.63	0.98	0.39	2
1015	1.12	0.99	1.04	1.05	0.14	2
1016	0.8	0.9	0.8	0.8	0.2	2
1017	1.2	1.3	1.0	1.2	0.4	2
1018	0.88	0.93	0.89	0.90	0.19	2
1020	0.90	0.90	0.90	0.90	0.16	2
1022	0.781	0.783	0.785	0.783	0.007	2
1023	0.59	0.60	0.66	0.62	30	n.r.
1024	n.r.	n.r.	n.r.	n.r.	n.r.	n.r.
1025	0.93	0.82	1.02	0.92	0.20	2
1027	0.85	0.85	0.82	0.84	0.11	2
1028	0.7	0.7	0.7	0.7	0.3	2
1029	0.95	0.87	0.80	0.87	0.16	2
1030	0.83	0.88	n.r.	0.83	0.21	2

n.r.: not reported
k.: coverage factor
Rep.: result of replicate analysis

Results reported by *OCLs* for the content of benzo[*a*]pyrene (BAP) in the cocoa butter test material. Assigned value is 0.86 µg/kg

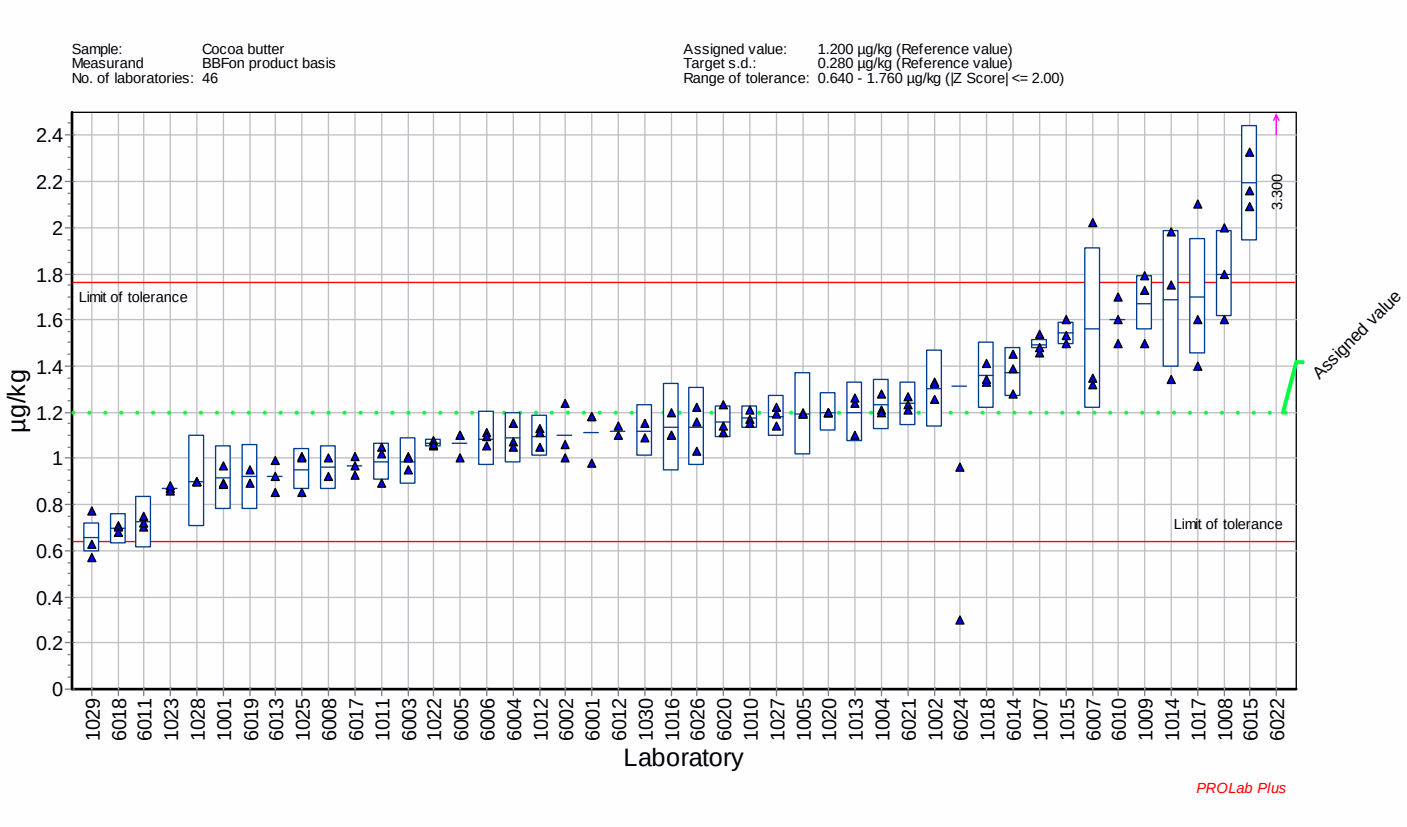
The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
6001	0.65	0.67	0.67	0.66	0.33	n.r.
6002	0.62	0.81	0.84	0.81	n.r.	n.r.
6003	0.95	0.98	0.96	0.96	0.19	2
6004	0.85	0.81	0.83	0.83	0.17	2
6005	1.2	1.1	1.1	1.1	25	2
6006	0.774	0.797	0.725	0.765	0.145	2
6007	1.01	0.89	0.89	0.93	0.40	2
6008	0.74	0.8	0.78	0.8	0.16	2
6010	1.9	1.9	1.9	1.9	n.r.	n.r.
6011	0.55	0.58	0.61	0.58	0.17	2
6012	0.84	0.83	0.79	0.82	20	n.r.
6013	0.726	0.670	0.772	0.722	35	56.9
6014	1.03	1.01	1.05	1.03	0.15	2
6015	0.68	0.67	0.65	0.67	0.15	2
6017	0.88	0.93	0.89	0.90	0.18	n.r.
6018	0.57	0.56	0.44	0.52	0.2	3
6019	0.69	0.25	n.r.	0.47	0.14	2
6020	0.93	0.97	0.90	0.92	0.08	2
6021	0.80	0.81	0.77	0.80	0.12	2
6022	0.6	0.6	1.2	<0.1	n.r.	n.r.
6024	1.58	0.63	1.66	1.29	n.r.	n.r.
6026	0.92	0.81	0.97	0.92	0.28	2

n.r.: not reported
k.: coverage factor
Rep.: result of replicate analysis

Distribution of individual results of replicate determinations reported for the benzo[*b*]fluoranthene (BBF) content of the cocoa butter test sample

blue triangles: individual results of replicate determinations, blue box: reported expanded measurement uncertainty (k=2), blue horizontal line in blue box: average of replicate determinations, green dotted line: assigned value, green area around assigned value: expanded uncertainty of the assigned value (k=2), red lines: lower and upper limit of satisfactory z-score range



Results reported by *NRLs* for the content of benzo[*b*]fluoranthene (BBF) in the cocoa butter test material. Assigned value is 1.20 µg/kg

The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
1001	0.895	0.886	0.968	0.92	0.28	2
1002	1.329	1.324	1.254	1.303	0.336	2
1004	1.28	1.21	1.20	1.23	0.22	2
1005	1.19	1.2	1.19	1.19	0.36	2
1007	1.54	1.46	1.48	1.49	0.04	2
1008	1.8	1.6	2.0	1.9	0.4	2
1009	1.79	1.73	1.5	1.67	0.23	2
1010	1.21	1.17	1.15	1.18	0.10	2
1011	0.89	1.05	1.02	0.99	0.16	2
1012	1.13	1.11	1.05	1.10	0.18	2
1013	1.10	1.24	1.26	1.20	0.26	2
1014	1.34	1.98	1.75	1.69	0.59	2
1015	1.50	1.60	1.53	1.54	0.10	2
1016	1.1	1.2	1.1	1.2	0.4	2
1017	2.1	1.6	1.4	1.7	0.5	2
1018	1.33	1.41	1.34	1.36	0.29	2
1020	1.20	1.20	1.20	1.20	0.17	2
1022	1.077	1.055	1.06	1.064	0.037	2
1023	0.87	0.86	0.88	0.87	30	n.r.
1024	n.r.	n.r.	n.r.	n.r.	n.r.	n.r.
1025	1.01	0.85	1.00	0.95	0.18	2
1027	1.22	1.14	1.19	1.18	0.18	2
1028	0.9	0.9	0.9	0.9	0.4	2
1029	0.63	0.57	0.77	0.66	0.13	2
1030	1.09	1.15	n.r.	1.09	0.22	2

n.r.: not reported
k.: coverage factor
Rep.: result of replicate analysis

Results reported by *OCLs* for the content of benzo[*b*]fluoranthene (BBF) in the cocoa butter test material. Assigned value is 1.20 µg/kg

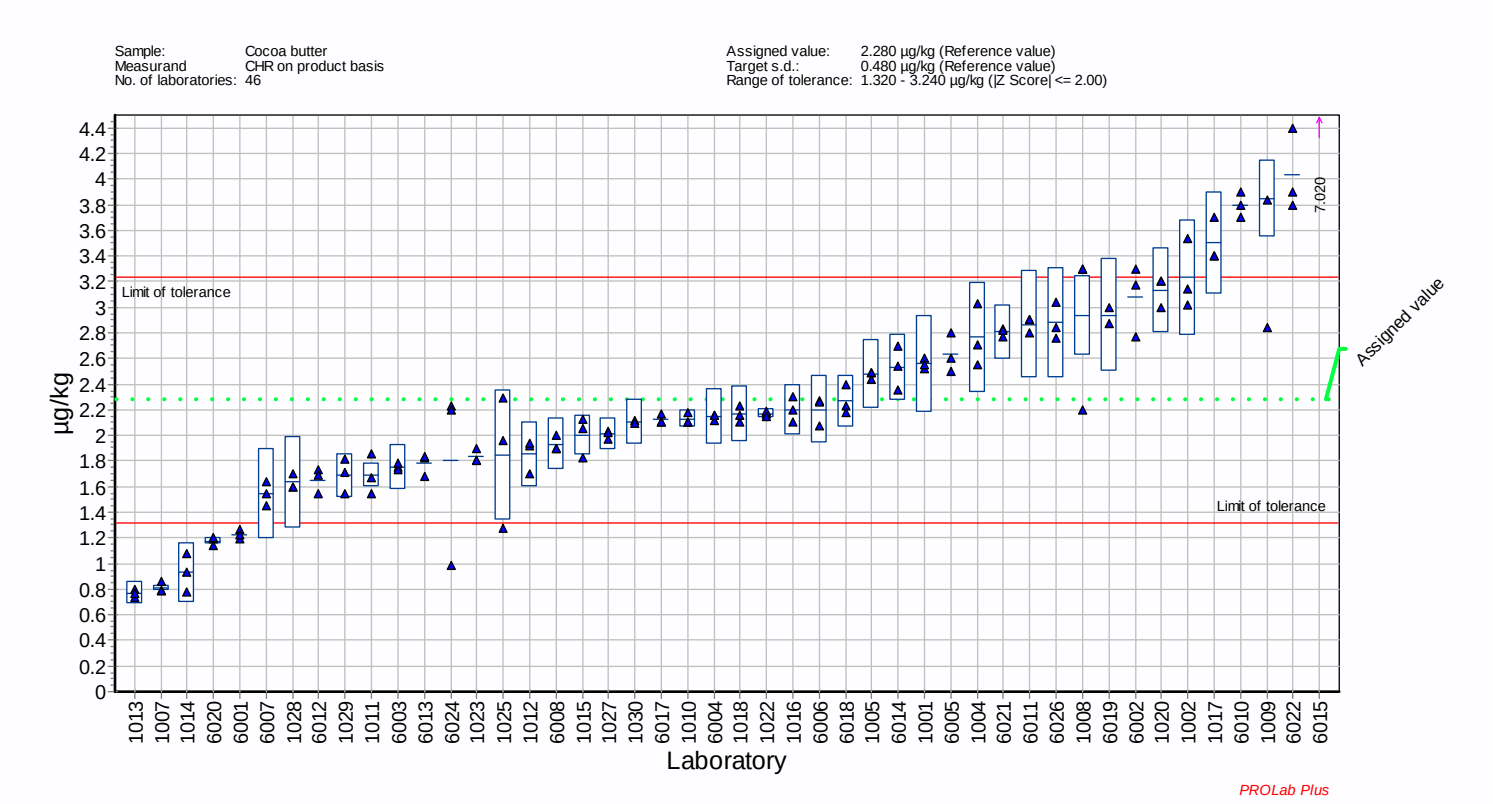
The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
6001	0.98	1.18	1.18	1.11	0.56	n.r.
6002	1.00	1.24	1.06	1.06	n.r.	n.r.
6003	1.01	0.95	1.00	0.99	0.20	2
6004	1.15	1.07	1.05	1.09	0.22	2
6005	1.1	1.1	1.0	1.1	25	2
6006	1.109	1.092	1.054	1.085	0.239	2
6007	2.02	1.35	1.32	1.56	0.70	2
6008	0.92	1.0	n.r.	1.0	0.2	2
6010	1.6	1.7	1.5	1.6	n.r.	n.r.
6011	0.75	0.70	0.72	0.72	0.22	2
6012	1.14	n.r.	1.10	1.08	20	n.r.
6013	0.924	0.854	0.991	0.923	35	56.9
6014	1.39	1.28	1.45	1.37	0.21	2
6015	2.33	2.16	2.09	2.19	0.50	2
6017	0.93	1.01	0.97	0.97	0.19	n.r.
6018	0.71	0.7	0.68	0.7	0.2	3
6019	0.95	0.89	n.r.	0.92	0.28	2
6020	1.23	1.14	1.11	1.18	0.14	2
6021	1.27	1.23	1.21	1.24	0.19	2
6022	2.9	3.8	3.2	3.3	0.25	n.r.
6024	<0.3	0.96	2.68	1.82	n.r.	n.r.
6026	1.16	1.03	1.22	1.16	0.35	2

n.r.: not reported
k.: coverage factor
Rep.: result of replicate analysis

Distribution of individual results of replicate determinations reported for the chrysene (CHR) content of the cocoa butter test sample

blue triangles: individual results of replicate determinations, blue box: reported expanded measurement uncertainty (k=2), blue horizontal line in blue box: average of replicate determinations, green dotted line: assigned value, green area around assigned value: expanded uncertainty of the assigned value (k=2), red lines: lower and upper limit of satisfactory z-score range



Results reported by *NRLs* for the content of chrysene (CHR) in the cocoa butter test material. Assigned value is 2.28 µg/kg

The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
1001	2.52	2.6	2.55	2.6	0.77	2
1002	3.532	3.02	3.139	3.230	0.895	2
1004	3.03	2.71	2.55	2.76	0.86	2
1005	2.49	2.49	2.44	2.47	0.54	2
1007	0.79	0.86	0.79	0.81	0.04	2
1008	3.3	2.2	3.3	3.3	0.7	2
1009	4.86	2.84	3.84	3.84	0.61	2
1010	2.18	2.11	2.1	2.14	0.14	2
1011	1.54	1.67	1.86	1.69	0.18	2
1012	1.92	1.94	1.7	1.85	0.51	2
1013	0.8	0.74	0.77	0.77	0.18	2
1014	1.08	0.93	0.78	0.93	0.47	2
1015	2.13	2.05	1.83	2.00	0.31	2
1016	2.1	2.2	2.3	2.2	0.4	2
1017	3.4	3.4	3.7	3.5	0.8	2
1018	2.11	2.23	2.16	2.17	0.44	2
1020	3.00	3.20	3.20	3.13	0.66	2
1022	2.169	2.191	2.147	2.169	0.071	2
1023	1.8	1.9	1.8	1.8	30	n.r.
1024	n.r.	n.r.	n.r.	n.r.	n.r.	n.r.
1025	2.29	1.28	1.96	1.84	1.02	2
1027	1.97	2.03	2.02	2.01	0.25	2
1028	1.6	1.6	1.7	1.6	0.7	2
1029	1.81	1.54	1.71	1.69	0.34	2
1030	2.12	2.09	n.r.	2.12	0.36	2

n.r.: not reported
k.: coverage factor
Rep.: result of replicate analysis

Results reported by *OCLs* for the content of chrysene (CHR) in the cocoa butter test material. Assigned value is 2.28 µg/kg

The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
6001	1.19	1.26	1.22	1.22	0.61	n.r.
6002	3.30	3.17	2.77	3.17	n.r.	n.r.
6003	1.75	1.73	1.78	1.75	0.35	2
6004	2.12	2.16	2.16	2.15	0.43	2
6005	2.5	2.8	2.6	2.6	25	2
6006	2.069	2.274	2.258	2.200	0.528	2
6007	1.64	1.54	1.45	1.54	0.70	2
6008	2	1.9	1.9	1.9	0.4	2
6010	3.8	3.9	3.7	3.8	n.r.	n.r.
6011	2.9	2.8	2.9	2.9	0.86	2
6012	1.73	1.69	1.54	1.65	20	n.r.
6013	1.82	1.68	1.84	1.78	35	56.9
6014	2.7	2.35	2.54	2.53	0.51	2
6015	6.86	6.87	7.33	7.02	1.50	2
6017	2.11	2.17	2.11	2.13	0.43	n.r.
6018	2.23	2.39	2.18	2.27	0.6	3
6019	3	2.87	n.r.	2.94	0.88	2
6020	1.19	1.2	1.14	1.17	0.06	2
6021	2.83	2.77	2.82	2.81	0.42	2
6022	3.8	4.4	3.9	4.0	0.20	n.r.
6024	2.23	0.98	2.2	1.80	n.r.	n.r.
6026	2.84	2.76	3.04	2.84	0.85	2

n.r.: not reported
k.: coverage factor
Rep.: result of replicate analysis

Results reported by *NRLs* for the sum of the four marker PAHs (SUM4PAH) in the cocoa butter test material. Assigned value is 5.89 µg/kg

Participant	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	
1001	5.67	0.96	2
1002	6.388	1.796	2
1004	6.65	0.78	2
1005	5.95	2.08	2
1007	4.46	0.07	2
1008	15.4	3.1	2
1009	8.78	1.36	2
1010	5.82	0.36	2
1011	4.63	0.60	2
1012	5.10	1.03	2
1013	3.91	0.42	2
1014	5.72	2.29	2
1015	6.49	0.47	2
1016	6.0	0.7	2
1017	7.8	1.1	2
1018	6.11	0.66	2
1020	6.80	0.86	2
1022	5.573	0.081	2
1023	4.9	20	n.r.
1024	7.84	1.38	2
1025	5.47	1.10	2
1027	5.71	0.41	2
1028	4.5	2.0	2
1029	3.84	0.70	2
1030	5.62	0.53	2

n.r.: not reported
k.: coverage factor

Results reported by *OCLs* for the sum of the four marker PAHs (SUM4PAH) in the cocoa butter test material. Assigned value is 5.89 µg/kg

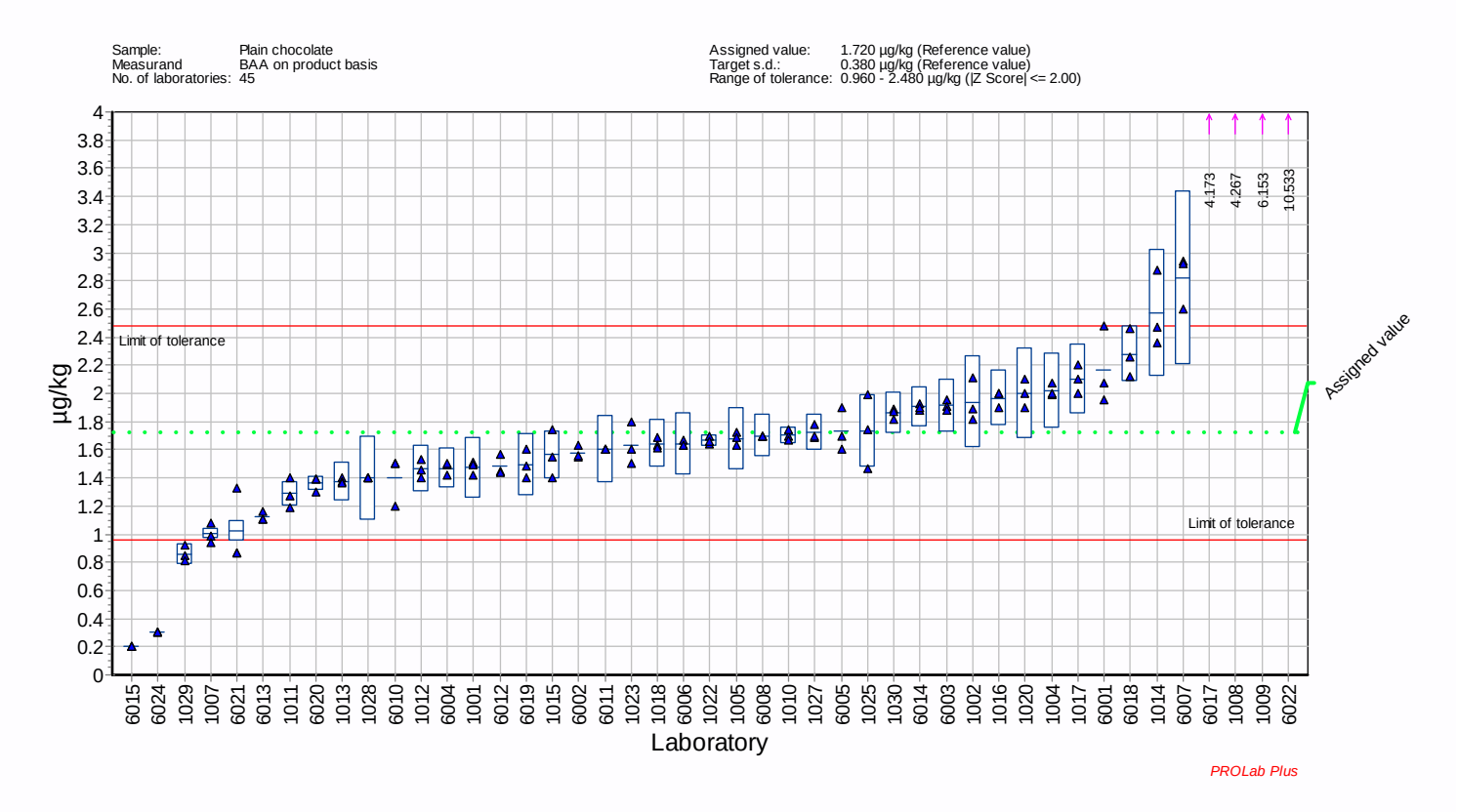
Participant	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	
6001	3.98	1.00	n.r.
6002	6.68	n.r.	n.r.
6003	5.37	0.55	2
6004	5.33	1.07	2
6005	6.2	25	2
6006	5.757	2.654	2
6007	5.53	2.40	2
6008	5.3	1.1	2
6010	8.5	n.r.	n.r.
6011	5.6	1.7	2
6012	4.89	30	n.r.
6013	4.68	35	n.r.
6014	6.74	1.35	2
6015	9.88	2.00	2
6017	8.01	1.60	n.r.
6018	5.84	1.0	3
6019	5.73	1.72	2
6020	4.45	0.29	2
6021	7.63	1.14	2
6022	14.6	0.90	n.r.
6024	6.82	n.r.	n.r.
6026	6.55	1.97	2

n.r.: not reported
k.: coverage factor

Distribution of individual results of replicate determinations reported for the benz[*a*]anthracene (BAA) content of the chocolate test sample.

Results are expressed on product basis (µg/kg chocolate).

blue triangles: individual results of replicate determinations, blue box: reported expanded measurement uncertainty (k=2), blue horizontal line in blue box: average of replicate determinations, green dotted line: assigned value, green area around assigned value: expanded uncertainty of the assigned value (k=2), red lines: lower and upper limit of satisfactory z-score range



Results reported by *NRLs* for the content of benz[*a*]anthracene (BAA) in the chocolate test material. Assigned value is 1.72 µg/kg

Results are expressed on product basis (µg/kg chocolate).

The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
1001	1.42	1.51	1.49	1.5	0.44	2
1002	1.887	1.813	2.112	1.047	0.352	2
1004	2.07	1.99	2.00	2.02	0.53	2
1005	1.63	1.72	1.69	1.68	0.44	2
1007	1.08	0.99	0.94	1.00	0.07	2
1008	4.1	4.5	4.2	2.4	0.5	2
1009	6.33	5.72	6.41	3.17	0.54	2
1010	1.70	1.67	1.74	1.70	0.12	2
1011	1.40	1.19	1.27	1.29	0.18	2
1012	1.53	1.40	1.46	1.46	0.33	2
1013	1.40	1.37	1.36	1.38	0.28	2
1014	2.88	2.36	2.47	2.57	0.9	2
1015	1.74	1.40	1.55	1.56	0.34	2
1016	1.9	2.0	2.0	2.0	0.4	2
1017	2.2	2.1	2.0	2.1	0.5	2
1018	1.69	1.63	1.61	1.64	0.34	2
1020	1.90	2.10	2.00	2.00	0.64	2
1022	1.642	1.694	1.661	1.665	0.084	2
1023	1.6	1.5	1.8	1.6	30	n.r.
1024	n.r.	n.r.	n.r.	n.r.	n.r.	n.r.
1025	1.74	1.99	1.47	1.73	0.52	2
1027	1.78	1.69	1.70	1.72	0.26	2
1028	1.4	1.4	1.4	1.4	0.6	2
1029	0.85	0.92	0.81	0.86	0.15	2
1030	1.89	1.82	1.87	1.82	0.29	2

n.r.: not reported

k.: coverage factor

Rep.: result of replicate analysis

Results reported by *OCLs* for the content of benz[*a*]anthracene (BAA) in the chocolate test material. Assigned value is 1.72 µg/kg

Results are expressed on product basis (µg/kg chocolate).

The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
6001	2.48	2.07	1.95	2.16	1.08	n.r.
6002	1.55	1.56	1.63	1.56	n.r.	n.r.
6003	1.95	1.91	1.88	1.91	0.38	2
6004	1.49	1.42	1.50	1.47	0.29	2
6005	1.6	1.7	1.9	1.7	25	2
6006	1.672	1.628	1.631	1.644	0.444	2
6007	2.94	2.60	2.92	1.60	0.70	2
6008	1.7	1.7	1.7	1.7	0.3	2
6010	1.5	1.5	1.2	1.4	n.r.	n.r.
6011	1.6	1.6	1.6	1.6	0.48	2
6012	1.57	1.45	1.44	1.48	20	n.r.
6013	1.11	1.11	1.16	1.13	35	n.r.
6014	1.88	1.93	1.90	3.44	0.52	2
6015	<0.2	<0.2	<0.2	<0.2	n.r.	n.r.
6017	4.12	4.22	4.18	4.17	0.83	n.r.
6018	2.26	2.46	2.12	2.28	0.6	3
6019	1.60	1.40	n.r.	2.95	0.89	2
6020	1.30	1.39	1.39	1.36	0.10	2
6021	1.33	0.87	0.87	1.02	0.15	2
6022	11.4	11.7	8.5	11.6	0.14	n.r.
6024	<0.3	<0.3	<0.3	<0.3	n.r.	n.r.
6026	n.r.	n.r.	n.r.	1.48	0.44	2

n.r.: not reported

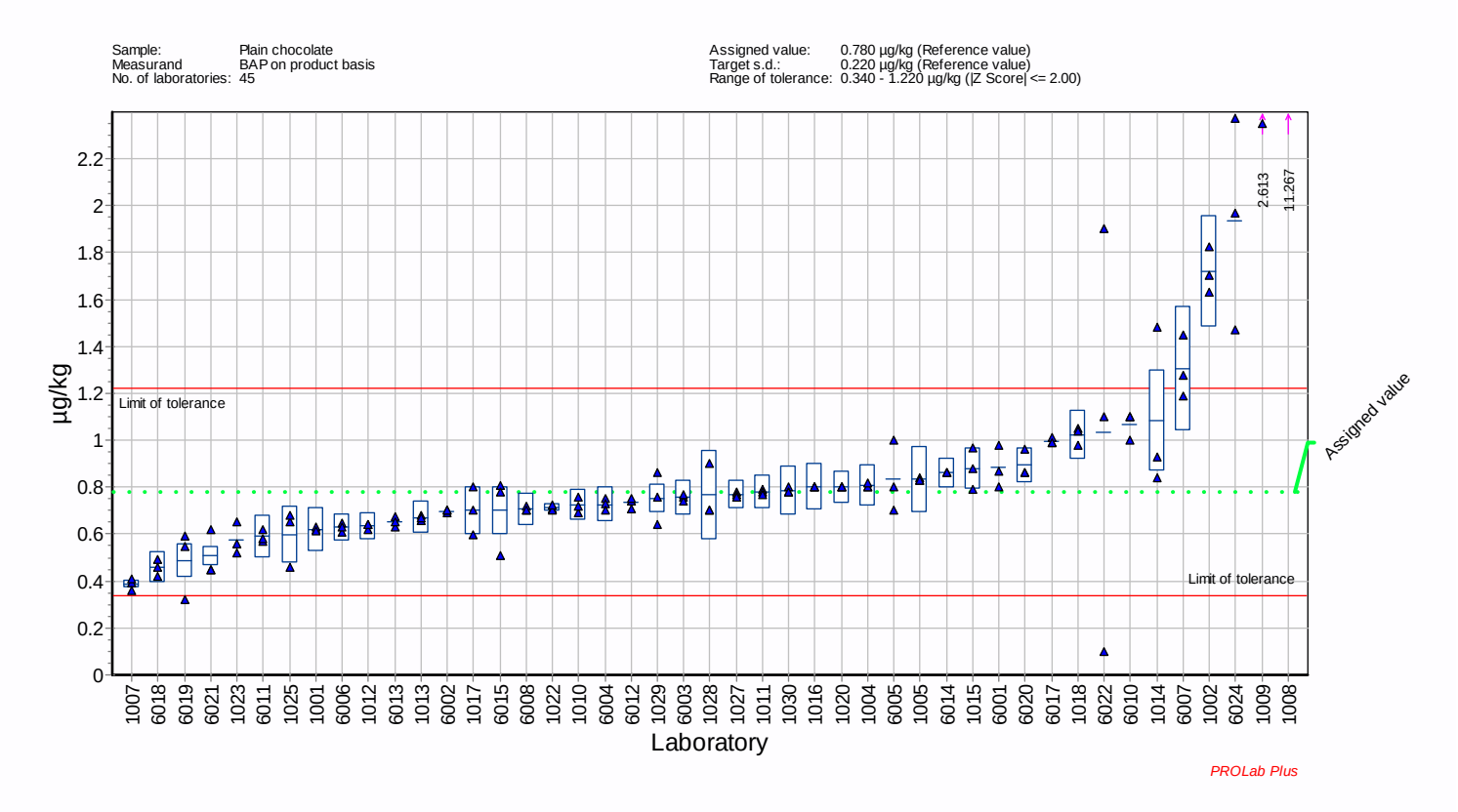
k.: coverage factor

Rep.: result of replicate analysis

Distribution of individual results of replicate determinations reported for the benzo[*a*]pyrene (BAP) content of the chocolate test sample.

Results are expressed on product basis (µg/kg chocolate)

blue triangles: individual results of replicate determinations, blue box: reported expanded measurement uncertainty (k=2), blue horizontal line in blue box: average of replicate determinations, green dotted line: assigned value, green area around assigned value: expanded uncertainty of the assigned value (k=2), red lines: lower and upper limit of satisfactory z-score range



Results reported by *NRLs* for the content of benzo[*a*]pyrene (BAP) in the chocolate test material. Assigned value is 0.78 µg/kg

Results are expressed on product basis (µg/kg chocolate).

The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
1001	0.618	0.630	0.612	0.62	0.19	2
1002	1.633	1.825	1.702	0.933	0.259	2
1004	0.80	0.80	0.82	0.81	0.18	2
1005	0.84	0.83	0.83	0.83	0.28	2
1007	0.39	0.41	0.36	0.39	0.03	2
1008	14.1	9.8	9.9	6.1	1.2	2
1009	2.35	2.69	2.80	1.35	0.20	2
1010	0.76	0.69	0.72	1.30	0.24	2
1011	0.79	0.78	0.77	0.78	0.14	2
1012	0.64	0.62	0.64	0.63	0.12	2
1013	0.68	0.66	0.67	0.67	0.14	2
1014	0.84	1.48	0.93	1.08	0.43	2
1015	0.97	0.79	0.88	0.88	0.18	2
1016	0.8	0.8	0.8	0.8	0.2	2
1017	0.6	0.7	0.8	0.7	0.2	2
1018	1.05	0.98	1.04	1.02	0.21	2
1020	0.80	0.80	0.80	0.80	0.14	2
1022	0.706	0.704	0.723	0.711	0.033	2
1023	0.56	0.52	0.65	0.58	30	n.r.
1024	n.r.	n.r.	n.r.	1.03	0.17	2
1025	0.65	0.68	0.46	0.59	0.24	2
1027	0.78	0.77	0.76	0.77	0.12	2
1028	0.9	0.7	0.7	0.8	0.4	2
1029	0.76	0.64	0.86	0.76	0.12	2
1030	0.8	0.78	0.78	0.78	0.21	2

n.r.: not reported

k.: coverage factor

Rep.: result of replicate analysis

Results reported by *OCLs* for the content of benzo[*a*]pyrene (BAP) in the cohocolate test material. Assigned value is 0.78 µg/kg

Results are expressed on product basis (µg/kg chocolate).

The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
6001	0.98	0.87	0.80	0.88	0.44	n.r.
6002	0.69	0.70	0.70	0.70	n.r.	n.r.
6003	0.76	0.74	0.77	0.76	0.15	2
6004	0.75	0.73	0.70	0.73	0.15	2
6005	0.7	0.8	1.0	0.8	25	2
6006	0.646	0.611	0.629	0.629	0.12	2
6007	1.45	1.19	1.28	0.74	0.30	2
6008	0.7	0.7	0.7	0.7	0.14	2
6010	1.1	1	1.1	1.1	n.r.	n.r.
6011	0.62	0.57	0.58	0.59	0.18	2
6012	0.74	0.75	0.71	0.73	20	n.r.
6013	0.674	0.632	0.652	0.653	35	n.r.
6014	0.86	0.86	0.86	0.86	0.13	2
6015	0.51	0.81	0.78	0.70	0.20	2
6017	0.99	1.01	0.99	1.00	0.2	n.r.
6018	0.49	0.46	0.42	0.46	0.2	3
6019	0.59	0.32	n.r.	0.44	0.13	2
6020	0.96	0.86	0.86	0.90	0.15	2
6021	0.62	0.45	0.45	0.51	0.08	2
6022	1.1	1.9	0.1	1.5	0.42	n.r.
6024	1.47	2.37	1.97	1.94	n.r.	n.r.
6026	n.r.	n.r.	n.r.	0.63	0.19	2

n.r.: not reported

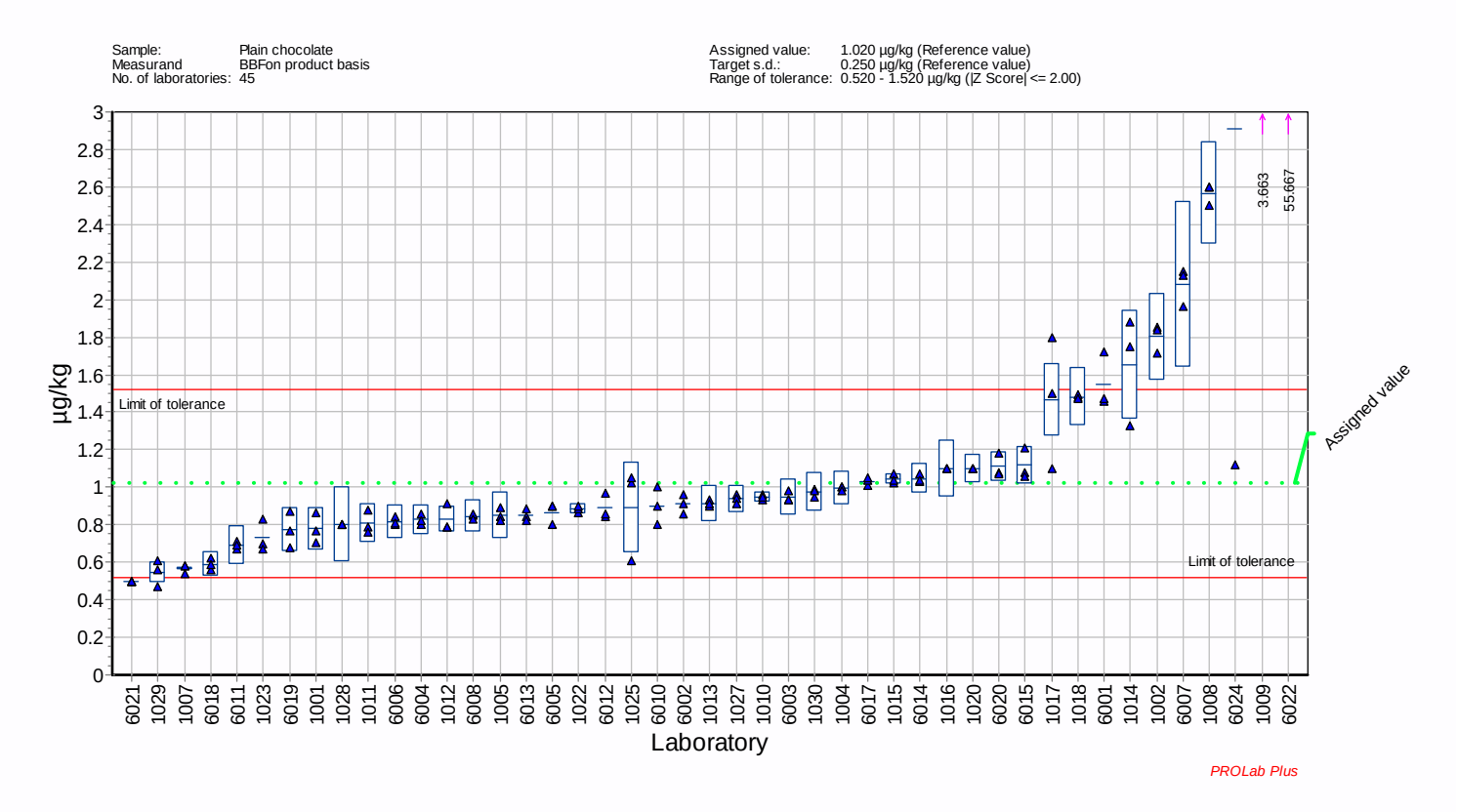
k.: coverage factor

Rep.: result of replicate analysis

Distribution of individual results of replicate determinations reported for the benzo[*b*]fluoranthene (BBF) content of the chocolate test sample.

Results are expressed on product basis (µg/kg chocolate)

blue triangles: individual results of replicate determinations, blue box: reported expanded measurement uncertainty (k=2), blue horizontal line in blue box: average of replicate determinations, green dotted line: assigned value, green area around assigned value: expanded uncertainty of the assigned value (k=2), red lines: lower and upper limit of satisfactory z-score range



Results reported by *NRLs* for the content of benzo[*b*]fluoranthene (BBF) in the chocolate test material. Assigned value is 1.02 µg/kg

Results are expressed on product basis (µg/kg chocolate).

The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
1001	0.708	0.862	0.768	0.78	0.23	2
1002	1.716	1.851	1.841	0.977	0.252	2
1004	1.00	1.00	0.98	0.99	0.18	2
1005	0.84	0.82	0.89	0.85	0.25	2
1007	0.58	0.58	0.54	0.57	0.02	2
1008	2.5	2.6	2.6	1.4	0.3	2
1009	3.47	3.56	3.96	1.89	0.26	2
1010	0.96	0.93	0.95	0.95	0.06	2
1011	0.88	0.79	0.76	0.81	0.21	2
1012	0.79	0.79	0.91	0.83	0.14	2
1013	0.90	0.93	0.91	0.91	0.19	2
1014	1.88	1.75	1.33	1.65	0.58	2
1015	1.07	1.02	1.04	1.04	0.05	2
1016	1.1	1.1	1.1	1.1	0.3	2
1017	1.5	1.8	1.1	1.5	0.4	2
1018	1.48	1.49	1.47	1.48	0.31	2
1020	1.10	1.10	1.10	1.10	0.15	2
1022	0.866	0.886	0.900	0.884	0.055	2
1023	0.70	0.67	0.83	0.73	30	n.r.
1024	n.r.	n.r.	n.r.	n.r.	n.r.	n.r.
1025	1.02	1.05	0.61	0.89	0.48	2
1027	0.96	0.94	0.91	0.94	0.14	2
1028	0.8	0.8	0.8	0.8	0.4	2
1029	0.61	0.56	0.47	0.55	0.11	2
1030	0.99	0.95	0.98	0.95	0.20	2

n.r.: not reported

k.: coverage factor

Rep.: result of replicate analysis

Results reported by *OCLs* for the content of benzo[*b*]fluoranthene (BBF) in the chocolate test material. Assigned value is 1.02 µg/kg

Results are expressed on product basis (µg/kg chocolate).

The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
6001	1.72	1.46	1.47	1.55	0.77	n.r.
6002	0.86	0.96	0.91	0.91	n.r.	n.r.
6003	0.98	0.93	0.93	0.95	0.19	2
6004	0.86	0.80	0.82	0.83	0.16	2
6005	0.9	0.9	0.8	0.9	25	2
6006	0.841	0.803	0.807	0.817	0.180	2
6007	2.15	2.13	1.96	1.18	0.50	2
6008	0.85	0.86	0.83	0.86	0.17	2
6010	0.9	0.8	1.0	0.9	n.r.	n.r.
6011	0.71	0.67	0.69	0.69	0.21	2
6012	0.84	0.86	0.97	0.89	20	n.r.
6013	0.844	0.822	0.885	0.850	35	n.r.
6014	1.03	1.07	1.04	1.90	0.29	2
6015	1.21	1.08	1.06	1.12	0.20	2
6017	1.01	1.04	1.05	1.03	0.21	n.r.
6018	0.56	0.62	0.59	0.59	0.2	3
6019	0.87	0.68	n.r.	1.51	0.45	2
6020	1.08	1.18	1.07	1.11	0.16	2
6021	<0.5	<0.5	<0.5	<0.5	n.r.	n.r.
6022	78.5	81.9	6.6	80.2	1.72	n.r.
6024	3.71	1.12	3.91	2.91	n.r.	n.r.
6026	n.r.	n.r.	n.r.	0.83	0.25	2

n.r.: not reported

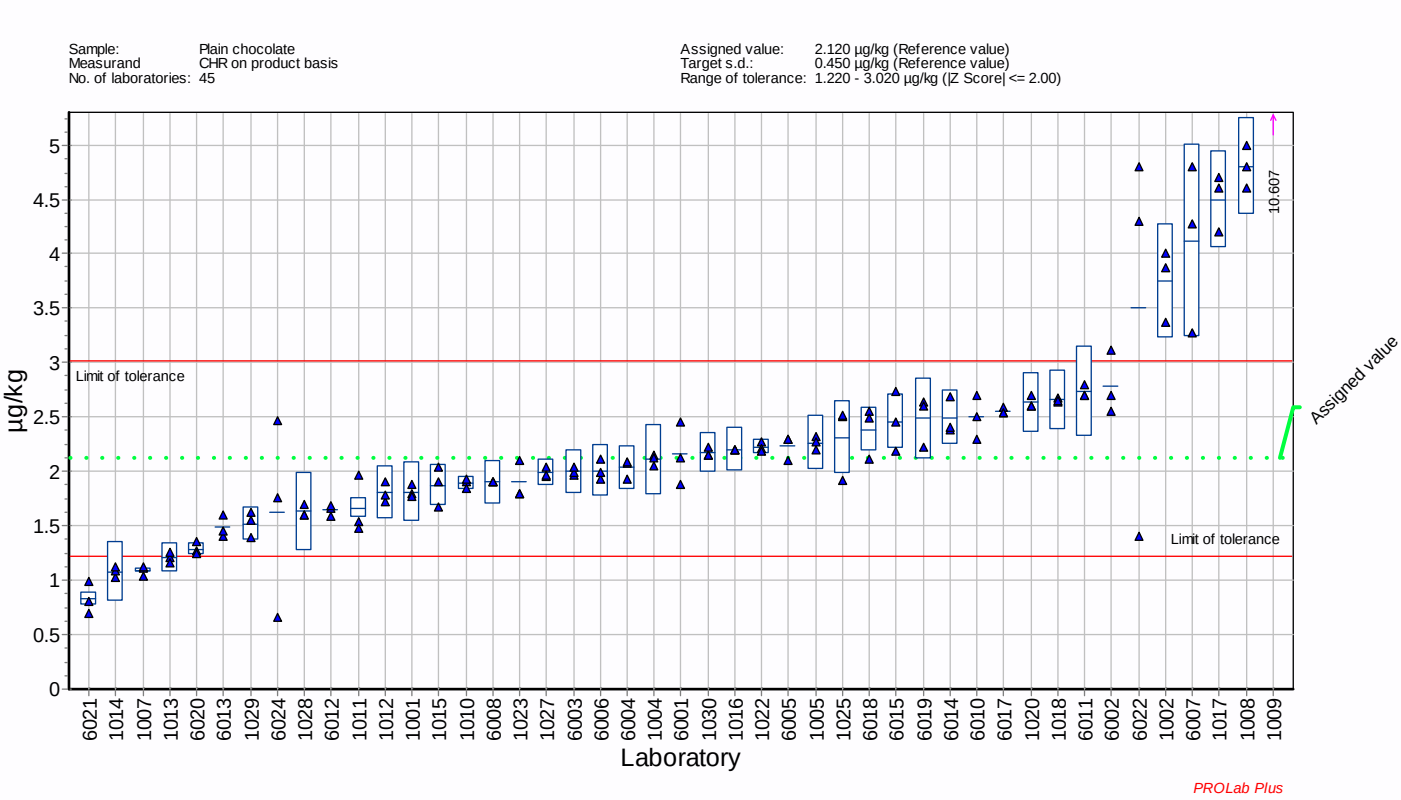
k.: coverage factor

Rep.: result of replicate analysis

Distribution of individual results of replicate determinations reported for the chrysene (CHR) content of the chocolate test sample.

Results are expressed on product basis (µg/kg chocolate).

blue triangles: individual results of replicate determinations, blue box: reported expanded measurement uncertainty (k=2), blue horizontal line in blue box: average of replicate determinations, green dotted line: assigned value, green area around assigned value: expanded uncertainty of the assigned value (k=2), red lines: lower and upper limit of satisfactory z-score range



Results reported by *NRLs* for the content of chrysene (CHR) in the chocolate test material. Assigned value is 2.12 µg/kg

Results are expressed on product basis (µg/kg chocolate).

The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
1001	1.79	1.88	1.77	1.80	0.54	2
1002	3.869	3.373	4.010	2.024	0.561	2
1004	2.15	2.12	2.05	2.11	0.66	2
1005	2.32	2.2	2.27	2.26	0.5	2
1007	1.11	1.12	1.04	1.09	0.04	2
1008	4.8	5.0	4.6	2.7	0.5	2
1009	12.32	9.35	10.15	5.46	0.87	2
1010	1.90	1.85	1.93	1.89	0.13	2
1011	1.97	1.54	1.48	1.66	0.18	2
1012	1.91	1.72	1.78	1.81	0.49	2
1013	1.21	1.16	1.26	1.21	0.26	2
1014	1.03	1.09	1.12	1.08	0.54	2
1015	1.90	1.67	2.04	1.87	0.38	2
1016	2.2	2.2	2.2	2.2	0.4	2
1017	4.2	4.7	4.6	4.5	0.9	2
1018	2.64	2.68	2.65	2.66	0.54	2
1020	2.60	2.70	2.60	2.63	0.55	2
1022	2.276	2.216	2.191	2.228	0.140	2
1023	1.8	1.8	2.1	1.9	30	n.r.
1024	n.r.	n.r.	n.r.	n.r.	n.r.	n.r.
1025	2.50	2.51	1.92	2.31	0.67	2
1027	2.04	1.96	1.97	1.99	0.25	2
1028	1.6	1.6	1.7	1.6	0.7	2
1029	1.55	1.39	1.62	1.52	0.30	2
1030	2.15	2.15	2.22	2.15	0.36	2

n.r.: not reported

k.: coverage factor

Rep.: result of replicate analysis

Results reported by *OCLs* for the content of chrysene (CHR) in the chocolate test material. Assigned value is 2.12 µg/kg

Results are expressed on product basis (µg/kg chocolate).

The uncertainty refers to the value for proficiency assessment.

Participant	Rep 1	Rep 2	Rep 3	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	µg/kg	µg/kg	µg/kg	
6001	2.46	1.88	2.13	2.16	1.08	n.r.
6002	2.55	3.12	2.70	2.70	n.r.	n.r.
6003	1.97	1.99	2.04	2.00	0.40	2
6004	1.93	2.08	2.09	2.03	0.41	2
6005	2.1	2.3	2.3	2.2	25	2
6006	2.110	1.924	1.987	2.007	0.482	2
6007	4.80	3.27	4.28	2.33	1.00	2
6008	1.9	1.9	1.9	1.9	0.4	2
6010	2.7	2.5	2.3	2.5	n.r.	n.r.
6011	2.8	2.7	2.7	2.7	0.82	2
6012	1.66	1.69	1.59	1.65	20	n.r.
6013	1.60	1.41	1.45	1.49	35	n.r.
6014	2.38	2.69	2.41	4.51	0.90	2
6015	2.18	2.45	2.74	2.46	0.50	2
6017	2.54	2.59	2.54	2.56	0.51	n.r.
6018	2.11	2.55	2.49	2.38	0.6	3
6019	2.64	2.22	n.r.	4.90	1.47	2
6020	1.27	1.24	1.35	1.30	0.11	2
6021	0.99	0.80	0.70	0.82	0.12	2
6022	4.3	4.8	1.4	4.5	0.24	n.r.
6024	0.66	1.76	2.47	1.63	n.r.	n.r.
6026	n.r.	n.r.	n.r.	2.22	0.67	2

n.r.: not reported

k.: coverage factor

Rep.: result of replicate analysis

Results reported by *NRLs* for the fat content and the sum of the four marker PAHs (SUM4PAH) in the chocolate test material.

A) Results are expressed on product basis (µg/kg chocolate): Assigned value is: 5.64 µg/kg.

B) Results are expressed on fat basis (µg/kg fat): Assigned value is: 9.57 µg/kg.

A)

Participant	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	
1001	4.68	0.758	2
1002	4.980	1.424	2
1004	5.93	0.69	2
1005	5.62	1.97	2
1007	3.05	0.14	2
1008	13.8	2.8	2
1009	11.86	1.84	2
1010	5.26	0.23	2
1011	4.54	0.50	2
1012	4.73	0.95	2
1013	4.17	0.45	2
1014	6.39	2.55	2
1015	5.35	0.54	2
1016	6.0	0.7	2
1017	8.7	1.2	2
1018	6.80	0.74	2
1020	6.53	0.87	2
1022	5.488	0.040	2
1023	4.8	20	n.r.
1024	n.r.	n.r.	n.r.
1025	5.53	1.88	2
1027	5.42	0.40	2
1028	4.6	2.1	2
1029	3.68	0.66	2
1030	5.70	0.54	2

B)

Participant	Fat content	Value for proficiency assessment	Uncertainty	k
	%	µg/kg	µg/kg	
1001	56.15	8.34	1.36	2
1002	54.129	9.210	2.633	2
1004	54.94	10.63	1.24	2
1005	52.89	10.63	3.72	2
1007	60.5	5.17	0.11	2
1008	56	21.6	4.3	2
1009	51.5	23.04	3.57	2
1010	55.38	9.50	0.41	2
1011	57.83	7.85	0.92	2
1012	56	8.41	1.69	2
1013	54.86	7.6	0.8	2
1014	58.5	10.90	4.37	2
1015	54.3	9.85	1.00	2
1016	54.2	11.1	1.4	2
1017	60.8	14.3	1.7	2
1018	54.7	12.43	1.33	2
1020	55.7	1.17	0.16	2
1022	53.9	10.192	0.742	2
1023	52	9.3	20	n.r.
1024	n.r.	n.r.	n.r.	n.r.
1025	49.96	11.3	4.11	2
1027	53	10.22	0.75	2
1028	55.4	8.2	3.4	2
1029	54.75	6.72	1.21	2
1030	54.09	10.55	1.00	2

n.r: not reported

k.: coverage factor

Results reported by *OCLs* for the fat content and the sum of the four marker PAHs (SUM4PAH) in the chocolate test material.

A) Results are expressed on product basis (µg/kg chocolate): Assigned value is: 5.64 µg/kg.

B) Results are expressed on fat basis (µg/kg fat): Assigned value is: 9.57 µg/kg.

A)

Participant	Value for proficiency assessment	Uncertainty	k
	µg/kg	µg/kg	
6001	6.76	3.38	n.r.
6002	5.87	n.r.	n.r.
6003	5.62	0.60	2
6004	5.06	1.00	2
6005	5.6	25	2
6006	5.096	2.345	2
6007	5.85	2.60	2
6008	5.2	1	2
6010	5.9	n.r.	n.r.
6011	5.6	1.7	2
6012	4.75	30	n.r.
6013	4.11	35	n.r.
6014	6.30	1.26	2
6015	4.27	0.90	2
6017	8.8	1.76	n.r.
6018	5.71	0.8	3
6019	5.14	1.54	2
6020	4.67	0.47	2
6021	2.73	0.41	2
6022	97.8	2.52	n.r.
6024	6.48	n.r.	n.r.
6026	5.16	1.55	2

B)

Participant	Fat content	Value for proficiency assessment	Uncertainty	k
	%	µg/kg	µg/kg	
6001	54.85	12.33	6.17	n.r.
6002	54.6	10.75	n.r.	n.r.
6003	55.1	10.19	1.09	2
6004	64.5	7.83	1.400	2
6005	n.r.	n.r.	n.r.	n.r.
6006	54.58	9.340	4.41	2
6007	57	10.33	4.60	2
6008	56	9.3	1.9	2
6010	55.8	10.6	n.r.	n.r.
6011	54.3	10.3	3.1	2
6012	57.3	8.84	30	n.r.
6013	54.8	7.5	35	n.r.
6014	55.2	11.4	2.30	2
6015	60.4	7.08	1.50	2
6017	54.74	16.0	3.20	n.r.
6018	54.9	10.4	1.5	3
6019	50.13	10.25	3.08	2
6020	54.6	8.55	0.86	2
6021	53.5	5.1	0.77	2
6022	55.5	176.2	4.55	n.r.
6024	55	11.78	n.r.	n.r.
6026	50	8.41	2.52	2

n.r: not reported

k.: coverage factor

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Abstract

This report presents the results of the tenth inter-laboratory comparison (ILC) organised by the European Union Reference Laboratory for Polycyclic Aromatic Hydrocarbons (EU-RL PAHs) on the determination of the four EU marker PAHs, benz[a]anthracene (BAA), benzo[a]pyrene (BAP), benzo[b]fluoranthene (BBF) and chrysene (CHR), in cocoa products, particularly in plain chocolate and cocoa butter. It was conducted under ISO Standard 17043 accreditation.
In agreement with National Reference Laboratories, the test material used in this exercise were commercial products.
Participants also received a solution of PAHs in solvent of their choice (either toluene or acetonitrile) with disclosed content for the verification of their instrument calibration.
Consensus values were used to benchmark the results reported by participants, as the experience of the analytical community with this analysis scope was considered not sufficient to provide reference values.
Both officially nominated National Reference Laboratories (NRLs) and official food control laboratories (OCLs) of the EU Member States were admitted as participants.

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