European Commission

JRC SCIENCE AND POLICY REPORTS

Report on the 2014 Proficiency Test of the European Union Reference Laboratory for Mycotoxins, for the Network of National Reference Laboratories

Determination of Aflatoxin B1 in Copra (Coconut powder)

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2014

Report EUR 26849 EN

Joint Research Centre

European Commission
Joint Research Centre
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JRC91908

EUR 26849 EN

ISBN 978-92-79-41181-6 (PDF)

ISSN 1831-9424(online)

doi:10.2787/98787

Luxembourg: Publications Office of the European Union, 2014

Abstract

This report presents the results of the PT of the EURL for Mycotoxins which focused on the determination of aflatoxin B1 (Afla B1) in coconut powder samples.
Sixty-one participants from 31 countries registered for the exercise and fifty-eight sets of results were reported.
Only z-scores were used for an evaluation of performance. In total 91 % of the attributed z scores were below an absolute value of two, which indicated that most of the participants performed satisfactorily.

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Determination of Aflatoxin B1 in Copra (Coconut powder)

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Project ID: AFLATOXIN B1 in COCONUT (Copra)
PT coordinator: Maciej Kujawski

September 2014

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1. Executive Summary

The Institute for Reference Materials and Measurements (IRMM) of the Joint Research Centre (JRC), a Directorate-General of the European Commission, operates the European Union Reference Laboratory (EURL) for Mycotoxins. One of its core tasks is to organise proficiency tests (PTs) among appointed National Reference Laboratories (NRLs).

This report presents the results of the PT on the determination of aflatoxin B1 (Afla B1) in coconut powder samples.

The test items were two naturally contaminated and one blank coconut powder. The materials were produced by the IRMM and dispatched to the participants in April 2014. Each participant received three sachets of approximately 50 g per test material.

Sixty-one participants from 31 countries registered for the exercise and fifty-eight sets of results were reported.

The assigned values, established by exact-matching double isotope dilution mass spectrometry at the EURL for Mycotoxins, were 5.76 µg/kg (Sample A) and 28.5 µg/kg (Sample B) for Afla B1. The expanded uncertainties of the respective assigned values were 0.23 and 1.5 µg/kg.

Participants were invited to report the uncertainty of their measurements. This was done by the majority of laboratories.

Laboratory results were rated with z-scores and zeta-scores in accordance with ISO 13528 and the International Harmonized Protocol for the Proficiency Testing of Analytical Chemistry Laboratories. The z-score compares the participant's deviation from the reference value with the target standard deviation accepted for the proficiency test, whereas the zeta-score provides an indication of whether the participant's estimate of uncertainty is consistent with the observed deviation from the assigned value.

Only z-scores were used for the evaluation of performance. In total 91 % of the attributed z-scores were below an absolute value of two, which indicated that most of the participants performed satisfactorily.

2. Introduction

Aflatoxins are mycotoxins that are found mainly in crops such as maize, cottonseed, peanuts and tree nuts, including coconut. They are produced by strains of *Aspergillus flavus, Aspergillus parasiticus* and *Aspergillus nomius*. *Aspergillus flavus* produces B aflatoxins only, while the other species produce both B and G aflatoxins.

Toxic effects of aflatoxins include carcinogenic, mutagenic, teratogenic, and immunosuppressive activity. Aflatoxin B1 [**Figure 1**] is the most potent hepatocarcinogen known in mammals and it is classified by the International Agency of Research on Cancer (IARC) as Group 1 carcinogen [**1**].

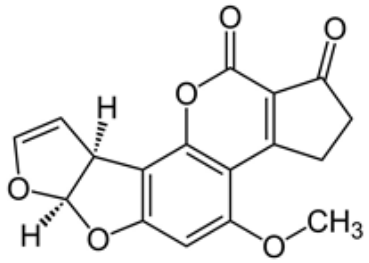


Figure 1: Chemical structures of aflatoxin B1

Commission Regulation (EC) No. 1881/2006 [**2**] lays down maximum limits of 5 µg/kg for Aflatoxin B1 in *Tree nuts, other than the tree nuts listed in 2.1.2 and 2.1.3, to be subjected to sorting, or other physical treatment, before human consumption or use as an ingredient in foodstuffs* (that includes coconut), and 2 µg/kg for Aflatoxin B1 in *Tree nuts, other than the tree nuts listed in 2.1.6 and 2.1.7, and processed products thereof, intended for direct human consumption or use as an ingredient in foodstuffs*. The European Commission also sets guideline limits for Afla B1 in animal feed in Commission Recommendations (2006/576/EC and 2002/32/EC) [**3-4**].

3. Scope

As stated in Article 32 of Regulation (EC) No 882/2004 [**5**], one of the core duties of the EURL is to organise proficiency tests (PTs) for the benefit of staff of NRLs. The scope of this PT was to test the competence of the appointed NRLs to determine the amount of Afla B1 in copra.

The PT design and data processing was in line with the International Harmonized Protocol for the Proficiency Testing of Analytical Chemical Laboratories [**6**].

The EURL Mycotoxins performed the assessment of the measurement results on the basis of requirements laid down in legislation and followed the administrative and logistic procedures of ISO 17043 [**7**]. JRC-IRMM is an ISO 17043 accredited PT provider.

3.1. Confidentiality

Confidentiality of the participants and their results towards third parties is guaranteed, with the exception to the customer requesting this PT, which is DG SANCO. Results were reported in a special online form for which each participant received an individual access code.

4. Time frame

The PT was initially announced by e-mail on the 5th of February 2014 and was published on the EURL web page [**8**]. The exercise was opened for registration on 10 March 2014 [**Annex 13.1**] and the deadline for

registration was 26th of March. The samples were dispatched to the participants on 8-9 April 2014 [**Annex 13.2**]. Reporting deadline was 6 June 2014.

5. Material

5.1. Preparation

The test materials used in this study were a blend of naturally contaminated copra with blank copra produced by cryo-milling to obtain a particle size < 500 µm. This material was further homogenised in a tumble mixer and was then packed in vacuum sealed aluminium sachets, taking portions from different places of the lot at random, and making up to a total sample size of ca. 50 g.

5.2. Homogeneity

To verify the homogeneity of the test materials 10 units each per material (Sample A and Sample B) were selected at random. Two independent determinations per unit were performed with a liquid-chromatography-fluorescence detection (LC-FLD) based method, which has been validated in-house. The order of measurements of the batch was randomised. Homogeneity was evaluated according to ISO 13528:2005 [**9**]. The material proved to be adequately homogeneous [**Annex 13.3**].

5.3. Stability

The stability study was conducted following an isochronous experimental design [**10**]. Based on previous experience -18 °C was chosen as reference temperature for sample storage.

Stability was evaluated according to the International Harmonized Protocol for the Proficiency Testing of Analytical Chemistry Laboratories [**6**].

The materials proved to be adequately stable at 4 °C for the period between dispatch and the deadline for submission of results.

5.4. Distribution

The test materials were dispatched in polystyrene boxes, containing cooling packs, on 8th and 9th of April 2014. The samples were mostly received within 24 hours after dispatch.

Each participant received:
a) two units containing approximately 50 g of test materials,
b) one blank sample,
c) an accompanying letter with instructions on sample handling and reporting [**Annex 13.2**],
d) a sample receipt form [**Annex 13.4**] and
e) a registration key for the reporting interface, and a lab code.

The materials were shipped in a manner that 4 °C was not exceeded. Upon arrival storage was required to be at -18 °C until the analysis was performed.

6. Instructions to participants

The laboratories were asked to report the recovery corrected value as well as their expanded measurement uncertainty in µg/kg and their coverage factor.

Results were reported in a special online form for which each participant received an individual access code. A specific questionnaire was attached to this online form. The questionnaire was intended to provide further

information on the measurements and the laboratories. A copy of the questionnaire is presented in **Annex 13.5**.
Participants received the information that the materials were shipped in cooling packs and that upon arrival the materials needed to be stored immediately at -18°C until the analysis is performed.

7. Reference values and their uncertainties

Assigned values and their uncertainties for the test samples were established by "Exact-matching Double Isotope Dilution Mass Spectrometry" at IRMM. This methodology is considered to be a primary ratio method with a direct link to SI units [**11**]. The assigned values were 5.76 µg/kg (Sample A) and 28.5 µg/kg (Sample B) for Afla B1. The uncertainties of the respective assigned values were 0.23 and 1.5 µg/kg.

8. Evaluation of results

8.1. General observations

Sixty-one laboratories participated in this PT: NRLs from twenty-nine Member States (two different NRLs for food and feed in eight Member States), two expert reference laboratories from 3rd countries, and 20 appointed Official Control Laboratories (OCLs) from 7 Member States. Fifty-eight sets of results (sample A and sample B) were reported.

All laboratories were free to use their method of choice. The technique most commonly used for the determination of aflatoxin B1 was high-performance liquid chromatography (HPLC) with fluorescence detection (81%). Mass selective (MS) detection and enzyme linked immunosorbent assays (ELISA) were also employed, but significantly less commonly.

8.2. Scores and evaluation criteria

Individual laboratory performance was assessed in terms of z and zeta (ζ) scores in accordance with ISO 13528 [**9**] and the International Harmonised Protocol [**6**].

$$z = \frac{x_{lab} - X_{ref}}{\sigma_p} \qquad \textbf{Equation 1.}$$

$$\zeta = \frac{x_{lab} - X_{ref}}{\sqrt{u^2_{lab} + u^2_{ref}}} \qquad \textbf{Equation 2.}$$

where:

x_{lab} is the measurement result reported by a participant
X_{ref} is the reference value (assigned value)
u_{lab} is the standard uncertainty reported by a participant
u_{ref} is the standard uncertainty of the reference value
σ_p is the standard deviation for proficiency assessment (target standard deviation)

The standard deviation for proficiency assessment σ_p (target standard deviation) was fixed by the PT coordinator using the Horwitz equation modified by Thompson [**12**] (for analyte concentrations < 120 ppb):

$$\sigma_p = 0.22 \cdot c \qquad \textbf{Equation 3.}$$

where:

c = concentration of the measurand (assigned value, $X_{ref,}$) expressed as a dimensionless mass ratio, e.g. 1 ppb = 10^{-9}, 1 ppm = 10^{-6}

The z-score compares the participant's deviation from the reference value with the target standard deviation accepted for the proficiency test, σ_p. The z-score is interpreted as:

$\|z\| \leq 2$	satisfactory result
$2 < \|z\| \leq 3$	questionable result
$\|z\| > 3$	unsatisfactory result

The zeta (ζ)-score provides an indication of whether the participant's estimate of uncertainty is consistent with the observed deviation from the assigned value. The ζ-score is the most relevant evaluation parameter, as it includes all parts of a measurement result, namely the expected value, its uncertainty as well as the uncertainty of the assigned values.

The interpretation of the zeta-score is similar to the interpretation of the z-score:

$\|\zeta\| \leq 2$	satisfactory result
$2 < \|\zeta\| \leq 3$	questionable result
$\|\zeta\| > 3$	unsatisfactory result

An unsatisfactory |ζ|-score might be due to an underestimation of the uncertainty, or to a large error causing a large deviation from the reference value, or to a combination of the two factors. A laboratory with an unsatisfactory |ζ|-score indicated an uncertainty which is not consistent with the laboratory's deviation from the reference value.

8.3. Laboratory results and scoring

Statistical evaluation of the results was performed using MS Excel.

The robust mean values and robust standard deviations were computed according to Algorithm A of ISO 13528 [**9**] by application of a MS Excel macro that was written by the Analytical Methods Committee of The Royal Society of Chemistry (AMC) [**13**].

IDMS was used to establish the assigned value. Both z-scoring and zeta-scoring were calculated for Afla B1, however only unsatisfactory z-scores will result in the request for corrective actions.

Summaries of the statistical evaluation for Afla B1 and test sample are presented in **Table 1.** The results as reported by the participants are summarised in **Table 2** together with the z-scores and zeta-scores.

Figures 2 and 3 provide the individual laboratory values and their uncertainty as reported.

Table 1: Summary statistics for aflatoxin B1

		Sample A	Sample B
Number of results		58	58
Range of results	µg/kg	0.57-11.83	3.12-50.84
Median of results of participants	µg/kg	5.80	27.15
Mean of results of participants	µg/kg	5.787	26.61
Robust mean of results of participants	µg/kg	5.85	27.0
Assigned value	µg/kg	**5.76**	**28.5**
Expanded uncertainty (k=2) of the assigned value	µg/kg	0.23	1.5
Robust standard deviation ($\hat{\sigma}$)	µg/kg	1.11	5.41
Target standard deviation (fitness for purpose)	µg/kg	1.27	6.27
Number (percentage) of results of |z| > 2.0		5 (9%)	5 (9%)
Number (percentage) of results of |ζ| > 2.0		16 (28%)	19 (33%)

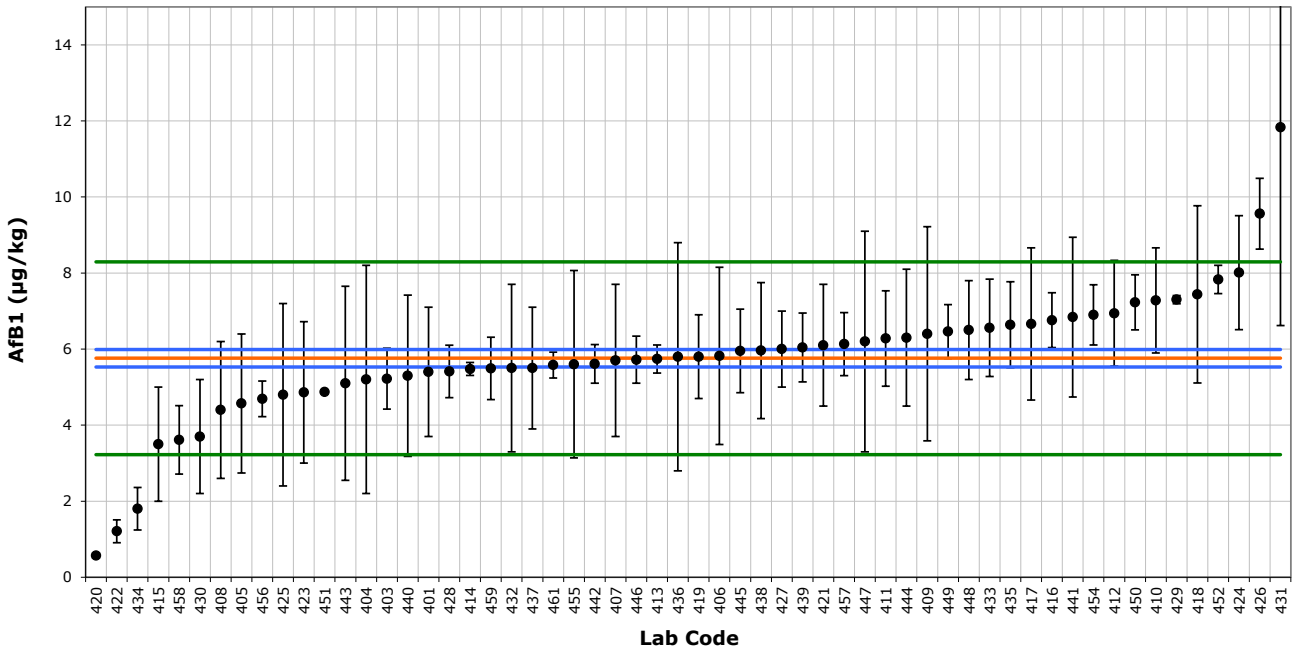
Table 2: Results of analysis, z-scores and zeta-scores for aflatoxin B1
(The meaning of colours: green – satisfactory, yellow – questionable, red – unsatisfactory)

Lab Code	SAMPLE A			SAMPLE B		
	Result [µg/kg]	z-score	zeta-score	Result [µg/kg]	z-score	zeta-score
401	5.4	-0.3	-0.4	27.4	-0.2	-0.2
402	*No result*			*No result*		
403	5.22	-0.4	-1.3	23.1	-0.9	-7.5
404	5.2	-0.4	-0.4	22.7	-0.9	-0.9
405	4.57	-0.9	-1.3	24.85	-0.6	-1.4
406	5.82	0.0	0.1	24.04	-0.7	-0.9
407	5.7	0.0	-0.1	34.2	0.9	0.9
408	4.4	-1.1	-1.5	21.3	-1.1	-1.7
409	6.4	0.5	0.5	31.3	0.4	0.4
410	7.28	1.2	2.2	29.42	0.1	0.3
411	6.279	0.4	0.8	28.692	0.0	0.1
412	6.94	0.9	1.7	29.6	0.2	0.4
413	5.74	0.0	-0.1	29.07	0.1	0.6
414	5.4771	-0.2	-2.0	22.5577	-0.9	-9.8
415	3.5	-1.8	-3.0	14.7	-2.2	-4.2
416	6.76	0.8	2.6	28.13	-0.1	-0.2
417	6.66	0.7	0.9	24.5	-0.6	-1.1
418	7.44	1.3	1.4	33.47	0.8	1.2
419	5.8	0.0	0.1	27.2	-0.2	-0.4
420	0.57	-4.1	-44.5	3.12	-4.0	-41.7
421	6.1	0.3	0.4	34.6	1.0	1.3
422	1.21	-3.6	-24.1	5.7	-3.6	-24.4
423	4.86	-0.7	-1.0	23.74	-0.8	-1.0
424	8.01	1.8	3.0	31.5	0.5	1.0
425	4.8	-0.8	-0.8	22.9	-0.9	-1.0
426	9.56	3.0	7.9	39.4	1.7	2.8
427	6	0.2	0.5	28.7	0.0	0.1
428	5.41	-0.3	-1.0	23.4	-0.8	-3.2
429	7.3	1.2	12.8	34.6	1.0	10.2
430	3.7	-1.6	-2.7	18	-1.7	-2.9
431	11.83	4.8	2.3	50.84	3.6	2.0
432	5.5	-0.2	-0.2	28.8	0.0	0.1
433	6.56	0.6	1.2	29.22	0.1	0.2
434	1.8	-3.1	-13.1	9.6	-3.0	-11.7
435	6.64	0.7	1.5	29.59	0.2	0.4

436	5.8	0.0	0.0	28.6	0.0	0.0
437	5.5	-0.2	-0.3	25.6	-0.5	-0.7
438	5.96	0.2	0.2	33.2	0.7	0.9
439	6.04	0.2	0.6	26.3	-0.4	-1.1
440	5.3	-0.4	-0.4	20.2	-1.3	-2.0
441	6.84	0.9	1.0	25.5	-0.5	-0.8
442	5.61	-0.1	-0.5	25.65	-0.5	-2.2
443	5.1	-0.5	-0.5	24.1	-0.7	-0.7
444	6.3	0.4	0.6	30.3	0.3	0.4
445	5.95	0.1	0.3	30.6	0.3	2.6
446	5.72	0.0	-0.1	26.28	-0.4	-1.5
447	6.2	0.3	0.3	29.6	0.2	0.2
448	6.5	0.6	1.1	32.8	0.7	1.1
449	6.46	0.6	1.9	28.34	0.0	-0.1
450	7.23	1.2	3.9	36.64	1.3	4.2
451	4.87	-0.7		26.16	-0.4	
452	7.83	1.6	9.5	21.4	-1.1	-11.3
453	*No result*			*No result*		
454	6.9	0.9	2.8	34.3	0.9	3.9
455	5.6	-0.1	-0.1	27.1	-0.2	-0.2
456	4.69	-0.8	-4.1	22	-1.0	-5.2
457	6.13	0.3	0.9	29.06	0.1	0.3
458	3.61	-1.7	-4.6	19.668	-1.4	-3.4
459	5.49	-0.2	-0.6	23.8	-0.7	-2.5
460	*No result*			*No result*		
461	5.58	-0.1	-0.9	26.5	-0.3	-2.0

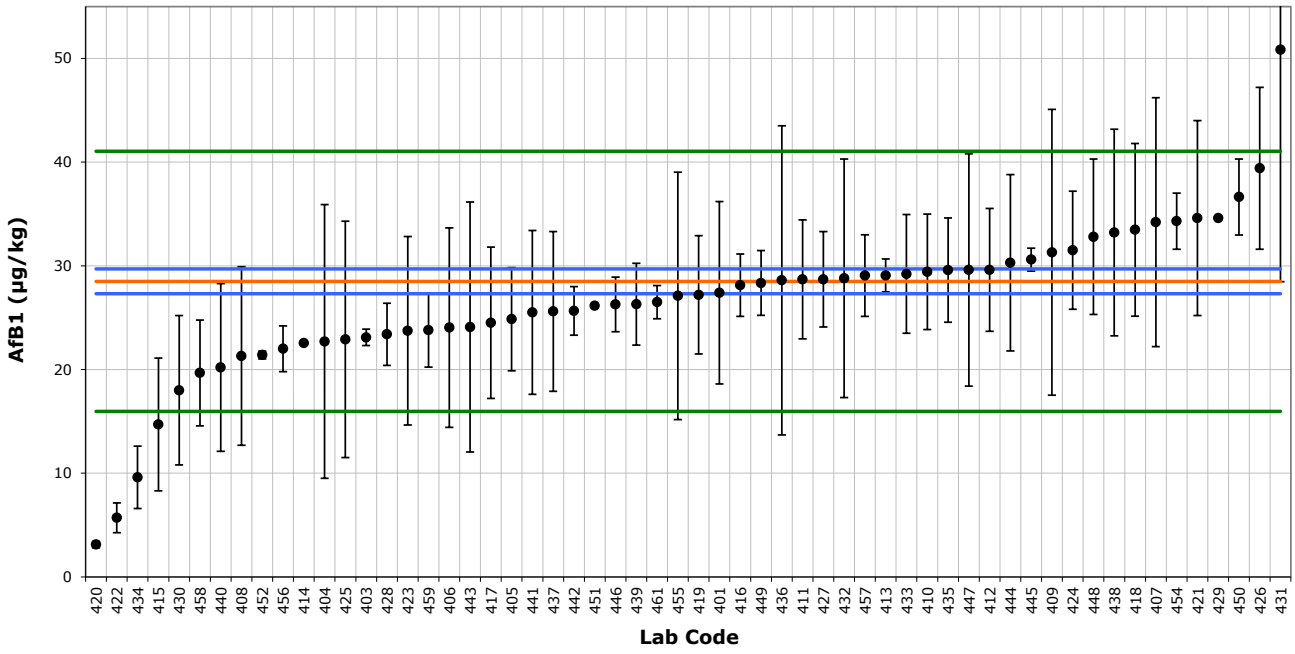
The results are written as reported by the laboratories.

Figure 2: EURL Mycotoxins PT 2014: Aflatoxin B1 in copra - Sample A
Certified value: Xref = 5.76 µg/kg; Uref = 0.23 µg/kg (k=2); σ = 1.267 µg/kg



This graph displays all revised measurement results and their associated uncertainties. The uncertainties are shown as reported.
The red line corresponds to **Xref**, the blue lines mark the boundary of the reference interval **(Xref ± 2uref)**, and the green lines that of the target interval **(Xref ± 2σ)**.

Figure 3: EURL Mycotoxins PT 2014: Aflatoxin B1 in copra - Sample B
Certified value: Xref = 28.5 µg/kg; Uref = 1.5 µg/kg (k=2); σ = 6.27 µg/kg



This graph displays all revised measurement results and their associated uncertainties. The uncertainties are shown as reported.
The red line corresponds to **Xref**, the blue lines mark the boundary of the reference interval **(Xref ± 2uref)**, and the green lines that of the target interval **(Xref ± 2σ)**.

8.4. Evaluation of the questionnaire

All laboratories that reported results, in total fifty-eight participants, supplied their filled in questionnaires. Experimental details along with a summary of the answers are presented in the **Annex 13.6.**

Most of the laboratories (81%) used HPLC-UV or FLD, whereas LC-MS and ELISA were used in 12% and 7% of cases, respectively. A majority used immunoaffinity columns for clean-up.

Most of the laboratories analysed annually 50-250 samples or more for Aflatoxin B1. Seventy-two percent of the participating laboratories are accredited for this type of analysis.

For the recovery estimation the two most commonly used methods were "internal standard added to sample" and spiking blank coconut sample.

Details about the applied methodologies – extraction, clean up, overnight stop, etc. - are presented in **Annex 13.6**. Details on the evaluation of various parameters asked in the questionnaire with respect to the z-scoring will be presented at the next EURL/NRL meeting in October 2014.

All participants found the instructions adequate and the registration-reporting interface the EU-RL received mostly positive feedback.

9. Conclusions

Fifty eight sets of results (Sample A and B) were reported for Afla B1.

Most of the participants performed satisfactory according the evaluation scheme used (target standard deviation computed according to Horwitz-Thompson). The consensus values and the values assigned by IDMS were in very good agreement. This underpins the validity and benefits of IDMS especially for smaller pools of participants during PT exercises.

10. Acknowledgements

The organizers of the study would like to thank Andreas Breidbach, Katy Kroeger-Negoita, Katrien Bouten, Stafanka Bratinova, Franz Ulberth and Beatriz de la Calle for their support.

The laboratories participating in this exercise, listed in [**Table 3**], are also kindly acknowledged.

Table 3: Participating laboratories

Organisation	Country
AGES GmbH	Austria
CODA-CERVA	Belgium
Bulgarian Food Safety Agency	Bulgaria
Institute of Public Health Dr. Andrija Štampar	Croatia
State General Laboratory	Cyprus
Department of Agriculture	Cyprus
Czech Agriculture and Food Inspection Authority (CAFIA)	Czech Republic
Central Institute for Supervising and Testing in Agriculture (UKZUZ)	Czech Republic
National Food Institute	Denmark
Danish Veterinary and Food Administration	Denmark
Agricultural Research Centre	Estonia
Finnish Customs Laboratory	Finland
Finnish Food Safety Authority Evira	Finland
Laboratoire SCL-35	France
Federal Inst f Risk Assessment BfR	Germany
General Chemical State laboratory	Greece
National Food Chain Safety Office, Food And Feed Safety Directorate	Hungary
Public Analyst's Laboratory, Dublin	Ireland
State Laboratory	Ireland
Istituto Superiore di Sanita'	Italy
ARPA Puglia	Italy
ASL della Provincia di VARESE	Italy
Azienda Sanitaria di Firenze	Italy
Institute of Food Safety, Animal Health and Environment "BIOR"	Latvia
National Food and Veterinary Risk Assessment Institute	Lithuania
Laboratoire National de santé	Luxembourg
Public Health Laboratory	Malta
RIKILT	Netherlands
NofaLab B.V.	Netherlands
National Veterinary Research Institute	Poland
ASAE	Portugal
Sanitary Veterinary and Food Safety Directorate Bucharest	Romania
Hygiene and Veterinary Public Health Institute Bucharest	Romania
Sanitary Veterinary and Food Safety Laboratory BRAILA	Romania
D.S.V.S.A. Cluj	Romania
DSVSA DOLJ	Romania
DSVSA CALARASI	Romania
DSVSA GALATI	Romania
Sanitary Veterinary Laboratory For Food Safety	Romania
Saudi Food And Drug Authority	Saudi Arabia
Saudi Food And Drug Authority	Saudi Arabia
Saudi Food And Drug Authority	Saudi Arabia
Health Sciences Authority	Singapore
State Veterinary And Food Institute, Veterinary and Food Institute in Košice	Slovakia
University of Ljubljana, Veterinary Faculty, National Veterinary Institute	Slovenia
National Laboratory For Health, Environment and Food	Slovenia
National Laboratory of Health, Environment and Food	Slovenia
National Center For Food	Spain
Govern de les Illes Balears	Spain
ainia	Spain
National Food Agency	Sweden
National Veterinary Institute (SVA)	Sweden
Kantonales Laboratorium Basel-Landschaft	Switzerland
Food & Environment Research Agency	United Kingdom
Kent County Council	United Kingdom
Staffordshire County Council	United Kingdom

Organisation	Country
Lancashire County Scientific Services	United Kingdom
Hampshire Scientific Service	United Kingdom

11. Abbreviations

Afla B1 (AfB1)	Aflatoxin B1
ANOVA	Analysis of variance
EC	European Commission
ELISA	Enzyme linked immunosorbent assay
EU	European Union
EURL	European Reference Laboratory
FLD	Fluorescent detection
HPLC	High-performance liquid chromatography
IAC	Immunoaffinity column
IDMS	Isotope Dilution Mass Spectrometry
IRMM	Institute for Reference Materials and Measurements
ISO	International Organisation for Standardisation
IUPAC	International Union for Pure and Applied Chemistry
JRC	Joint Research Centre
LOD	Limit of Detection
LOQ	Limit of Quantification
NRL	National Reference Laboratory
OCL	Official Control Laboratory
PT	Proficiency Test

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[**2**] Commission Regulation (EC) No 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs
http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=CONSLEG:2006R1881:20100701:EN:PDF

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[**4**] Commission Directive 2002/32/EC of 7 May 2002 on undesirable substances in animal feed
http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=CONSLEG:2002L0032:20100302:EN:PDF

[**5**] Commission 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
http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=CONSLEG:2004R0882:20060525:EN:PDF

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[**12**] Thompson, M., Recent trends in inter-laboratory precision at ppb and sub-ppb concentrations in relation to fitness for purpose criteria in proficiency testing, Analyst, 2000, 125, 385-386

[**13**] Analytical Methods Committee, Robust statistics: a method of coping with outliers, Technical brief No 6, Apr 2001.
http://www.rsc.org/pdf/amc/brief6.pdf

13. Annexes

13.1. Opening of registration

EUROPEAN COMMISSION
DIRECTORATE-GENERAL
JOINT RESEARCH CENTRE
Directorate D - Institute for Reference Materials and Measurements

European Union Reference Laboratory for Mycotoxins

Geel, 28 March 2014

Interlaboratory Comparison of the EU-RL for Mycotoxins

Dear Sir/Madam,

On behalf of the EU-RL for Mycotoxins, I announce the opening of the interlaboratory comparison for the determination of aflatoxin B_1 in coconut powder (not defatted).

This proficiency test (PT) was announced by e-mail on the 5th February 2014.

More details on the PT design will be communicated upon sample dispatch.

The EU-RL Mycotoxins would like to inform you that, according to Regulation (EC) No 882/2004, the participation of activities organised by the EU-RL is mandatory for the National Reference Laboratories.

- For NRLs the participation is mandatory and therefore free of charge.
- The participation fee for official food control laboratories is 270 Euro per participant. The full participation fee is payable upon dispatch of the test samples. The IRMM will contact participants with details of the payment.

Confidentiality of the participants and results are guaranteed.

Registration will be possible from 10th March 2014 until midnight 26th March 2014

Dispatch of the PT materials is scheduled for the 1st and 2nd of April2014, so that the samples are supposed to arrive at your laboratory latest on Friday 4th of April 2014.

In order to register, laboratories must:

1. Enter the details online by using this link: PT EU-RL 2014 AFLATOXIN Registration LINK

   (you will find a guide how to register as attachment to this email)

Retieseweg 111, B-2440 Geel - Belgium. Telephone: (32-14) 571 211
Telephone: direct line (32-14) 571 229. Fax: (32-14) 571 783.

E-mail: irc-irmm-eurl-mycotox@ec.europa.eu
Web site: http://irmm.jrc.ec.europa.eu

2. Print the completed form (approved and confirmed version) when the system asks to do so, sign it and stamp it with your company stamp

3. Send it to the PT coordinator of the EU-RL Mycotoxins indicated below:

Carsten MISCHKE

Tel: +32 14 573 011
Fax: +32 14 571 783

Email: JRC-IRMM-EURL-MYCOTOX@ec.europa.eu

Deadline for reporting will be the 23rd May 2014. You will receive the link for entering the results upon reception of the PT samples.

A detailed outline of the PT together with your laboratory code will accompany the PT sample parcel. Please do not hesitate to contact us if you require further clarification.

For all types of communication please contact us at the e-mail address:

JRC-IRMM-EURL-MYCOTOX@ec.europa.eu

With kind regards,

Carsten MISCHKE

(on behalf of the Operating Manager of the EU-RL Mycotoxins)

Cc: Frans Verstraete, Franz Ulberth, Beatriz De La Calle, Joerg Stroka

Retieseweg 111, B-2440 Geel - Belgium. Telephone: (32-14) 571 211
Telephone: direct line (32-14) 571 229. Fax: (32-14) 571 783.

E-mail: irc-irmm-eurl-mycotox@ec.europa.eu
Web site: http://irmm.jrc.ec.europa.eu

13.2. Accompanying letter

EUROPEAN COMMISSION
JOINT RESEARCH CENTRE

Institute for Reference Materials and Measurements
European Union Reference Laboratory for Mycotoxins

Geel, 7 April 2014

Ref: EURL mycotoxins 2014 Proficiency Test of National Reference Laboratories (NRLs) and official control laboratories (OCLs) on aflatoxin B1 in copra/coconut

Dear Participant,

Please read the following information carefully before starting any analysis. If there are additional questions, do not hesitate to contact us by either phone or email (see details below).

The 2014 PT aims to:

Assess the content in two naturally contaminated test samples (marked as "Level A" and "Level B") and in addition an aflatoxin B1 free sample (marked as "Blank") which you may use to determine the recovery of your analysis. You will be asked to report the **recovery corrected value** (µg/kg), including your **recovery** (%) and **measurement uncertainty** (µg/kg) for a coverage factor of 2 (k=2).

The materials are shipped between cool packs; storage however should be at -18° C until the analysis is performed. A short period of 1-2 days without cooling is no harm for the material, but a longer period of storage above -18° C shall be avoided.

IMPORTANT NOTE: We would like to inform you that due to the production process (cryo-milling) the material resulted in very fine powder. As the material has been packed under vacuum the powder tends to clog. This must be considered during extraction. In our own experiments we have observed that simple shaking might not be suitable and we recommend high speed blending for extraction.

Please confirm the parcel's receipt by fax or e-mail immediately, by using the "**Materials receipt form**". If any material is damaged, please request new material immediately.

Please report all requested results and answer the questionnaire at
https://web.jrc.ec.europa.eu/ilcReportingWeb
The password key for this interface is included in the parcel with the test materials.
When you enter the password key please pay attention to the capital letters!

Print the final pdf and return the signed and stamped report sheet NOT later than **30th May 2014** to:

Carsten Mischke
FAX: +32-14-571 783
E-mail: JRC-IRMM-EURL-Mycotox@ec.europa.eu

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

With kind regards,

Carsten Mischke
(on behalf of the Operating Manager of the EU-RL Mycotoxins)

Retieseweg 111, B-2440 Geel - Belgium. Telephone: (32-14) 571 211. http://irmm.jrc.ec.europa.eu
Telephone: direct line (32-14) 571 229. Fax: (32-14) 571 783.

E-mail: jrc-irmm-eurl-mycotox@ec.europa.eu

13.3. Homogeneity test

Homogeneity according to ISO 13528:2005 [15]	**Sample A**	**Sample B**
	Afla B1	Afla B1
Mean	4.702	22.536
$\hat{\sigma}$	1.034 (22%)	4.958 (22%)
0.3 $\hat{\sigma}$ (critical value)	0.310	1.487
S_X (standard deviation of sample averages)	0.296	1.427
S_W (within-sample standard deviation)	0.321	1.603
S_S (between-sample standard deviation)	0.190	0.867
S_S < 0.3 $\hat{\sigma}$	**Passed**	**Passed**

13.4. Acknowledgement of receipt form

EUROPEAN COMMISSION
JOINT RESEARCH CENTRE

Institute for Reference Materials and Measurements
European Union Reference Laboratory for Mycotoxins

Geel, 07 April 2014

PROFICIENCY TESTING MATERIALS RECEIPT FORM

Name:
Institute:
Address:
Member State:

NOTE: STORE ALL MATERIALS IN A FREEZER AT -18 °C!

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

Date of receipt	
All items have been received undamaged	YES / NO
If NO, please list damaged items:	

Contents of the parcel:

a) A bag including 3 test materials for analysis:
- Level A
- Level B
- Blank

b) A bag containing the following documents:
- This materials **receipt** form
- Copy of **instructions**
- Your **password** key to enter results & Laboratory **code** for your laboratory
- Draft paper version of the **questionnaire**

Please sign this completed form to and fax or e-mail it to:

Carsten MISCHKE
FAX: +32-14-571 783

E-mail: JRC-IRMM-CRL-MYCOTOX@ec.europa.eu

Retieseweg 111, B-2440 Geel - Belgium. Telephone: (32-14) 571 211
Telephone: direct line (32-14) 571 229. Fax: (32-14) 571 783.

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

Your Signature / Stamp here:

13.5. Questionnaire

Milc questionnaire

Comparison for AFLATOXIN B1 in COCONUT

Dear Participant, Please fill in your results and answer the questions. Print the final pdf and return the signed and stamped copy by fax +32 14 571 783 or by e-mail to JRC-IRMM-CRL-MYCOTOX@ec.europa.eu Thank you very much!

Submission Form

1. **How many samples does your laboratory analyse for Aflatoxin B1 per year?** *
- ◯ a) < 50 samples per year
- ◯ b) 50-250 samples per year
- ◯ c) 250-1000 samples per year
- ◯ d) more than 1000 samples per year

2. **Which food or feed matrices does your laboratory analyse for Aflatoxin B1 on a routine basis the most? (maximum 3)** *

3. **Are you accredited for the determination of Aflatoxin B1?** *
- ◯ a) Yes
- ◯ b) No

3.1. If YES, please specify the scope exactly how it is mentioned in your accreditation (e.g. Aflatoxin B1 +B2+G1+G2 in roasted peanuts by RPLC-FLD in the range of 0.1-10 µg/kg OR Aflatoxins in food etc.) *

4. **Proficiency test samples: COPRA / COCONUT**

4.1. Please indicate your recovery (%) *

4.2. Please indicate the LOD for Aflatoxin B1 of the method used (µg/kg). *

4.3. Please indicate the LOQ for Aflatoxin B1 of the method used (µg/kg). *

5. **How did you perform the recovery estimate?** *
- ◯ a) Using a CRM?
- ◯ b) Internal Standard to Sample
- ◯ c) Internal Standard to Extract
- ◯ d) International literature
- ◯ e) Isotope dilution
- ◯ f) Other (please specify)

5.1. If other please specify! *

6. **If you used a spiking solution to determine recovery, please mention the solvent of the solution and the concentration of aflatoxin B1 per mL?**

7. **Did you check your calibrant by UV-spectrophotometer?**
- ◯ a) Yes
- ◯ b) No

8. **During the analysis did you need to include any over night stop?** *
- ◯ a) Yes
- ◯ b) No

8.1. If YES, please state for which samples and at what stage of the analysis. *

9. **What was the extraction solvent used?** *

10. **What was the extraction mode (e.g. blending or sonication)?** *

11. **What was the extraction time?** *

12. **What was the solvent to sample ratio used during extraction (in mL/g)?** *

13. **Were any extraction aids added (e.g. NaCl)?** *

- ◯ a) Yes
- ◯ b) No

13.1. If YES, please state what and in which quantity. *

14. **What type of clean up methodology was used (e.g. immunoaffinity column)?** *

15. **If you used immunoaffinity columns...**

15.1. ... please specify the manufacturer of the immunoaffinity columns you used during the analysis!

15.2. ... please specify the brand and the production code of the immunoaffinity column!

16. **What type of detection method did you use?** *

- ◯ a) HPLC-FLD
- ◯ b) HPLC-UV
- ◯ c) LC-MS
- ◯ d) GC-MS
- ◯ e) Other (please specify)

16.1. If HPLC-FLD, please specify your method (type of column, mobile phase etc.)! *

16.2. If HPLC-UV, please specify your method (type of column, mobile phase etc.)! *

16.3. If LC-MS, please specify your method (type of column, ion information, mass fraction of sample injected [mg] etc.)! *

16.4. If GC-MS, please specify your method (type of column, split ratio, temperature (gradient) etc.)! *

16.5. If other, please specify your method!

17. **Which derivatisation method was applied (e.g. Kobra cell, OPA)** *

18. **How did you integrate the signals?**

- ◯ Automatic
- ◯ Manual

18.1. If automatic, did you confirm the integration correctness visually? *

- ◯ a) Yes
- ◯ b) No

19. **Did you use acid washed glassware?** *

- ◯ a) Yes
- ◯ b) No

20. **Was protection against daylight applied?** *

- ◯ a) Yes
- ◯ b) No

21. **Did you encounter any problems during the analysis?** *

- ◯ a) Yes
- ◯ b) No

21.1. If YES, what were the specific problems and to which samples do they apply? *

22. **Did you notice any unusual observations which, however, did not seem to have any effect on the results?** *

- ◯ a) Yes
- ◯ b) No

22.1. If YES, what were these observations and to which samples do they apply? *

23. **Did you find the instructions distributed for this PT adequate?** *

- ◯ a) Yes
- ◯ b) No

23.1. If NO, which parts do you think can improve? *

24. **What is your opinion about the registering / reporting format of this interface?**

25. **Any other comments you wish to address?**

13.6. Experimental details

Results and method performance characteristics for Aflatoxin B1

Lab Code	Technique	Sample A		Sample B		Coverage factor	Recovery [%]	LOD [µg/kg]	LOQ [µg/kg]
		Result [µg/kg]	Uncertainty [µg/kg]	Result [µg/kg]	Uncertainty [µg/kg]				
401	HPLC-FLD	5.4	1.7	27.4	8.8	2	93	0.2	0.02
403	HPLC-FLD	5.22	0.8	23.1	0.8	2	103.9	0.3	0.3
404	HPLC-FLD	5.2	3	22.7	13.2	2	97.2	0,02	0,1
405	HPLC-FLD	4.57	1.828	24.85	4.97	2	89	0.02	0.05
406	LC-MSMS	5.82	2.33	24.04	9.62	2	120	1	2.5
407	HPLC-FLD	5.7	2	34.2	12	2	83	0.05	0.10
408	LC-MS/MS	4.4	1.8	21.3	8.6	2	100	0.5	1
409		6.4	2.8	31.3	13.8	2			
410	photometrically at 450 nm	7.28	1.38	29.42	5.57	2	90.5	0.19	0.38
411	HPLC-FLD	6.279	1.256	28.692	5.738	2	85.8 for level 4µg/kg; 94.9 for level 30µg/kg	0.033	0.1
412	ELISA	6.94	1.39	29.6	5.92	2	98	1.08	1.44
413	HPLC-FLD	5.74	0.37	29.07	1.58	2	82.3	0.1	0.3
414	HPLC-FLD	5.4771	0.171688	22.5577	0.171688	2	86.48	0.23	0.77
415	HPLC-FLD	3.5	1.5	14.7	6.4	2	83	0.4	0.4
416	HPLC-FLD	6.76	0.72	28.13	3	2	92	0.2	0.4
417	HPLC-FLD	6.66	2	24.5	7.3	2	67	0.2	0.6
418		7.44	2.33	33.47	8.32	2			
419	HPLC-FLD	5.8	1.1	27.2	5.7	2	94.9 and 100.0	0.2	0.6
420	HPLC-FLD	0.57	0.06	3.12	0.31	3	65	2	0.5
421		6.1	1.6	34.6	9.4	2			
422	HPLC-FLD	1.21	0.3	5.7	1.43	2	87	0.6	1.0
423	HPLC-FLD	4.86	1.86	23.74	9.08	2	95.65	0.1	0.2
424	ELISA	8.01	1.5	31.5	5.7	2	80	0.3	0.6
425		4.8	2.4	22.9	11.4	2			
426		9.56	0.93	39.4	7.8	2			
427	HPLC-FLD	6	1	28.7	4.6	2	96.0	Not applicable	0.2
428		5.41	0.69	23.4	3	2			
429	HPLC-FLD	7.3	0.11	34.6	0.11	3	60	0.03	0.1
430	HPLC-FLD	3.7	1.5	18	7.2	2	77	0.03	0.06
431		11.83	5.21	50.84	22.37	2			
432	HPLC-FLD	5.5	2.2	28.8	11.5	2	107	0.1	0.2
433	HPLC-FLD	6.56	1.28	29.22	5.72	2	75 for sample A and 85 for sample B	0,2	0,24
434		1.8	0.56	9.6	3	2			
435	HPLC-FLD	6.64	1.13	29.59	5.03	2	87.21	0.2	0.6
436	HPLC-FLD	5.8	3	28.6	14.9	2	85	0.05	0.8
437	HPLC-FLD	5.5	1.6	25.6	7.7	2	110	0.1	0.3
438	HPLC-FLD	5.96	1.79	33.2	10.0	2	51.9	0.5	2.0

Lab Code	Technique	Sample A		Sample B		Coverage factor	Recovery [%]	LOD [µg/kg]	LOQ [µg/kg]
		Result [µg/kg]	Uncertainty [µg/kg]	Result [µg/kg]	Uncertainty [µg/kg]				
439	LC-MS	6.04	0.91	26.3	3.9	2	96.5	0.5	1
440	LC-MS	5.3	2.12	20.2	8.08	2	70	0.2	0.5
441	HPLC-FLD	6.84	2.1	25.5	7.9	2	Level A - 75; Level B - 93	0.04	0.2
442		5.61	0.51	25.65	2.34	2			
443	LC-MS	5.1	2.6	24.1	12.1	2	80	0.1	0.2
444	HPLC-FLD	6.3	1.8	30.3	8.5	2	92	0.06	0.2
445		5.95	1.1	30.6	1.1	2			
446		5.72	0.62	26.28	2.63	2			
447	HPLC-FLD	6.2	2.9	29.6	11.2	2	75	0.02	0.05
448	HPLC-FLD	6.5	1.3	32.8	7.5	2	88.34	0.2	0.6
449	Elisa method	6.46	0.71	28.34	3.12	2	71	1.01	2.02
450	HPLC-FLD	7.23	0.72	36.64	3.66	2	106.3	0.35	0.49
451	HPLC-FLD	4.87	0	26.16	0	2	98.88	0.02	0.07
452	HPLC-FLD	7.83	0.372	21.4	0.372	2	93.25	0.14	
454	LC-MS	6.9	0.79	34.3	2.7	2	109	0.2	0.6
455	HPLC-FLD	5.6	2.5	27.1	11.9	2	86	0.05	0.2
456	HPLC-FLD	4.69	0.469	22	2.2	2	92	0.1	0.1
457	HPLC-FLD	6.13	0.83	29.06	3.93	2	90	0.05	0.2
458	HPLC-FLD	3.61	0.9	19.668	5.1	2	93.7	0.5	0.5
459	HPLC-FLD	5.49	0.82	23.8	3.57	2	79	0.06	0.18
461	HPLC-FLD	5.58	0.34	26.5	1.6	2	90	0.5	1

How did you perform the recovery estimation?
If you used spiking solution to determine recovery, please mention the solvent of the solution and the concentration of aflatoxin B1 per mL.
Do you check your calibrant by UV-spectrophotometer?

Lab Code	Recovery estimation	Spiking solvent and concentration (Afla B1 per mL)	Calibrant UV-check?
401	Internal Standard to Sample	0.594 µg/ml in MeOH	Yes
403	Using a CRM		No
404	Internal Standard to Sample	0,25 µg B1/ml acetonitrile	No
405	Internal Standard to Extract	acetonitrile; 2.04 µg/ml	No
406	Using a CRM		No
407	standard addition to a blank sample	AcCN 2.01 ug/mL	No
408	A blank reference sample is spiked with a known amount of toxin, the IS is added after completion of the extraction	200 ng/ml in acetonitril	No
409			
410	fortified (spiking) sample	methanol; 1	Yes
411	Internal Standard to Sample	Acetonitril, Concentration for Aflatoxin B1 2.04 µg per ml	No
412	Internal Standard to Sample	Methanol, 70%, 0.010 µg/ml, by level 10 µg/kg.	Yes
413	Standard addition to blank sample	100 ppb (ng/mL) in ACN	No
414	Using a CRM		No
415	By spiked matrix	Acetonitrile, 200 ug/ml	No
416	Using a CRM		Yes
417	Internal Standard to Sample	Methanol-water (1:1), 40.2 ng/mL and 201 ng/mL	No
418			
419	Internal Standard to Sample	Toluen/ACN 9.84 ug/mL	Yes
420	Internal Standard to Extract	MeOH/H2O (80/20), 2 µg/kg	No
421			
422	Internal Standard to Sample	acetonitrile 0.5 ug/ml	No
423	Using a CRM	100 ng in methanol	No
424	Internal standard to blank	Methanol 70%	No
425			
426			
427	Using the "Blank" sample provided; spiking at 2ng/g.	200ng/mL spiking solution in methanol.	Yes
428			
429	Standard addition to the blank and to coco nut powder	methanol : water = 4:1 and 200 ng Aflatoxin B1	Yes
430	Internal Standard to Sample	toluen/ACN 98/2, conc: 1.34 ug/ml	Yes
431			
432	Internal Standard to Sample	23 ng/ml in methanol-water (40-60)	No
433	External standard to sample	Methanol 250 ng/ml	No
434			
435	Using a CRM	B1 concentration 100 µg/L, in acetonitrile	No
436	Internal Standard to Sample	200 ng/ml in methanol	Yes
437	Spiking on sample	Spiking solution in toluen CH3CN 1µg/ml 125 µl on 25g sample	Yes
438	Using a CRM		No
439	AFB1 standard to blank sample (coconut)	solvent: ACN, 1 ng/mL	No
440	Spiking solution to blank sample.	Acetonitril, 100 ng/mL.	No
441	spiking standard solution to blank sample	Methanol - water (1:1 v/v) c=1.0 ug/ml	Yes
442			
443	Internal Standard to Extract	blank extract, 80%MeOH, different conc levels 2, 12.5, 25, 40, 50 ug/l in final solution	No
444	Internal Standard to Sample	0.377 µg aflatoxin B1 in methanol	Yes
445			
446			
447	FAPAS sample		No
448	Spiking of the blank matrix 112	Methanol. Concentration per mL of B1 100,08 ng/mL	No
449	Using a CRM	-	Yes
450	Spiking solution	methanol , 0.0011958 ug /ml	No
451	Internal Standard to Sample	acetonitrile, 250 ng/mL, mixed certified solution	No
452	Internal Standard to Sample	215.28 ng/ml in chloroform	Yes
454	We used spiked samples	Spiking solution of B1 0.125 ng/µl in acetonitrile	Yes
455	External spiking solution to blank sample	Acetonitrile, 2.5 µg/mL	Yes
456	Internal Standard to Sample		No

Lab Code	Recovery estimation	Spiking solvent and concentration (Afla B1 per mL)	Calibrant UV-check?
457	Internal Standard to Sample	200 ng/ml in methanol	Yes
458	spiked sample	methanol, 18 ng/mL	Yes
459	standard addition to Extract		No
461	Internal Standard to Sample	5.025 ng	true

How many samples does your laboratory analyse for aflatoxin B1 per year?
Which food or feed matrices does your laboratory analyse most frequently for Aflatoxin B1 on a routine basis?
Are you accredited for the determination of aflatoxin B1? If yes, please specify the scope.

Lab Code	Samples annually	Matrices	Accredited?	Scope of accreditation
401	250-1000	Soya, nuts, figs	Yes	Cereal and cereal-based products, vegetables and fruits, complete feeds, feed materials excl cereals, nuts: 0,2-100 µg/kg, U=32%
403	< 50	Nuts, spices, animal feeding stuffs	Yes	Aflatoxins B1, G1, B2 and G2 using HPLC with fluorescence detection
404	< 50	nuts, dried fruit	Yes	Aflatoxin B1+B2+G1+G2 in food in the range of 0,1-150 µg/kg
405	250-1000	mixed feed, maize, nuts	Yes	Foodstuffs-Determination of aflatoxin B1 and the total of aflatoxins B1, B2, and G2 in cereals, nuts and derived products-High-performance liquid chromatographic method
406	50-250	feed material, compound feed	No	
407	50-250	Feed, maize and maize flour, pistachio	Yes	AfB1 and sum of AfB1+AfB2+AfG1+AfG2 in hazelnuts, peanuts, pistachio, figs and paprika powder by HPLC-FLD in the range of 0.10-3,40 (peanuts) 0.10-7.94 (pistachio)
408	< 50	Corn & nuts (mainly peanuts and pistachio nuts)	Yes	The determination in cereals, breakfast cereals, bread, pasta and nuts with LC-MS/MS of the following mycotoxins: DON, AFB1, AFB2, AFG1, AFG2, HT2, T2, ZEN, OTA, FB1, FB2 & FB3. Note (1): the range is not mentionned in the scope but the method has been validated using spikes between 2.5 and 7.5 ng/g, Note (2) Our scope doesn't include feed (for animals)
409				
410	50-250	feed cereals; complete feedingstuffs; nuts.	Yes	Aflatoxin B1 in feed and food by ELISA
411	250-1000	Nuts, cereals, spices	Yes	Aflatoxin B1+B2+G1+G2 in nuts , cereals, spices and their processed products in range of 0.1-50 µg/kg
412	50-250	Cereals.	Yes	Aflatoxin B1 in cereals and feed.
413	50-250	figs, peanut, babyfood	Yes	Aflatoxin B1,Afatoxin B2, Aflatoxin G1, Aflatoxin G2 in food of plant origin by HPLC
414	50-250	nuts /cereals/drived	No	
415	50-250	Cereals, dried fruits	Yes	Aflatoxin B1 and Sum of Aflatoxins B1+B2+G1+G2 in cereals, dried fruits, nuts and spices by HPLC-FLD , post column derivatization and immunoaffinity clean-up
416	50-250	cereals, feed, nuts	Yes	Aflatoxin B1+B2+G1+G2 in cereals and products derived from cereals by HPLC-FLD and Aflatoxin B1 in feed
417	50-250	Feed material, feedingstuffs	Yes	AFB1, feed material, feedingstuffs, 0.2-30 ug/kg
418				
419	50-250	cereals and their products, dried fruits, animal feed	Yes	Determinação das aflatoxinas B1, B2, G1 e G2. Cromatografia líquida de alta resolução (HPLC) em Frutos secos e produtos à sua base, oleoginosas, cereais e produtos à sua base, leguminosas, coco e especiarias e alimentação animal
420	250-1000	Cereals, nuts, animal feed	Yes	Aflatoxins in food from cereals, aflatoxins in feed
421				
422	< 50	Feed	No	
423	250-1000	nuts, feedstuff, grain	Yes	Aflatoxin B1, B2, G1, G2 in grain, maize, nuts, pistachios, figs, herbs, spices, nut butter, feedstuff and other products by RPLC-FLD
424	250-1000	dried fruits, cereals, feed materials	Yes	Aflatoxin B1 in cereals by ELISA
425				
426				
427	50-250	Cereals, Spices, Nuts	Yes	The determination of aflatoxins B1, B2, G1 and G2 in food by immunoaffinity column extraction and HPLC. Scope: Seeds, cereals, nut products, dried fruit and dried fruit products 0.2 - 20µg/kg; Shelled nuts 0.2 - 25µg/kg; Nuts and groundnuts in shell 0.2 - 40µg/kg; Spices 0.2 - 30µg/kg; Babyfood 0.05 - 20µg/kg (B1 only).
428				
429	50-250	Spices and cereals	Yes	C-P-215 Aflatoxine B & G und Ochratoxin A mittels HPLC-FLD und Coring-Zelle
430	< 50	figs, pistacios, peanuts	Yes	Aflatoxin B1+B2+G1+G2 in roasted peanuts, fiigs, pistacios, Brazil nuts, paprika by RPLC-FLD
431				
432	250-1000	nuts, baby food, spices	Yes	Aflatoxin B1, B2, G1 and G2 in food
433	50-250	cereals, nuts, spices	Yes	Aflatoxin B1 and Aflatoxin B1+B2+G1+G2 in nonanimal products food

Lab Code	Samples annually	Matrices	Accredited?	Scope of accreditation
434				
435	50-250	peanuts, oil seeds, cereals,spices	Yes	aflatoxins in food
436	50-250	Figs, Peanuts, Almonds	No	
437	250-1000	oilseeds, spices, feed	Yes	AF B1 in animal feed (SPE, HPLC);Oilseeds :peanut, pistachios, hazelnut. Figs. Paprika AF B1+B2+G1+G2 (IAC, HPLC)
438	< 50	Feed	No	
439	50-250	cereals, silage, complete feed	Yes	Multiresidual method for the determination of mycotoxins in feed by LC-MS
440	50-250	Nuts, cereals, dried fruit.	Yes	Aflatoxins B1, B2, G1 and G2 in Tree nuts, Ground nuts Almond, Pistachio, Peanuts by LC-MS/MS in the range of 0.5-10 µg/kg.
441	50-250	CEREALS, CEREAL PRODUCTS, FEEDINGSTUFFS	Yes	Aflatoxin B1+B2+G1+G2 in cereals, cereals products, nuts,feedingstuffs etc. by HPLC/FL with post column derivatization inthe range 0.2-10 ug/kg
442				
443	50-250	cereals and milled products, figs, nuts, spices, cofee...	Yes	aflatoxin in cereals and milled products, nuts and dried fruts by LC-MS in range 0.2-1000ug/kg and in infant cereals in range 0.06-1000ug/kg
444	250-1000	maize, cotton seed, maize byproducts	Yes	Determination of Aflatoxin B1 in animal feeding stuffs
445				
446				
447	50-250	cereals, nuts, dried fruit, spices	Yes	Aflatoxin B1, B2, G1, G2 in Food
448	250-1000	Dried nuts, Dried Fruits, Cereals	Yes	Aflatoxin Determination B1, B2, G1, G2 in dried nuts, dried fruits, cereals and their products and of Aflatoxin B1 in baby foods based on cereals with HPLC
449	50-250	Cereals and processed products, oilseeds.	Yes	Aflatoxin B1 in vegetables products.
450	50-250	nuts, pistachio, almonds	Yes	Aflatoxin B1, B2, G1, G2 in nuts
451	< 50	pistachios, peanuts	Yes	Aflatoxin B1+B2+G1+G2 in pistachios and other matrices by HPLC-FLD in the range of 0.1-5 µg/kg
452	50-250	Nuts, Spices and seeds	Yes	Determination of individual and total aflatoxins in nuts, nut products, cereals and seeds
454	250-1000	CERIALS, NUTS, DRIED FRUITS AND THEIR PRODUCTS	Yes	Determination of Mycotoxins (B1,B2,G1,G2,T-2,HT-2,ZON,OA,FB1,FB2,DON) in nuts ,cerials and their products by LC-MS/MS in the range of 0.5- 15µg/kg
455	250-1000	Cereal, infant food, animal feed	Yes	Aflatoxins B1,B2,G1,G2 using immunoaffinity clean-up and HPLC in foods, beverages, baby foods, cereals, cereal products, plant materials and plant extracts
456	50-250	raw material, feed mixture for cows and pigs	Yes	Aflatoxin B1 in Feed
457	< 50	cereals, product cereals, complementary feedstuffs	Yes	Aflatoxin B1 and total aflatoxins by HPLC/FLD
458	more than 1000	nuts, dried fruits, cereals	Yes	Aflatoxin B1, B2, G1, G2 and sum by HPLC FLD in nuts, dried fruits, cereals, spices, in the range of 0.5 and 24 µg/Kg
459	50-250	nuts, cereals, feeds	Yes	Aflatoxins B1, B2, G1, G2 in feeds, animal tissues, cereals, rice, cereal products, oilseeds, nuts, leguminous plants, processed fruits and vegetables, spices and figs by HPLC/FLD in range 0.08 - 100 ug/kg.
461	50-250	nuts, dried fruit	Yes	Aflatoxin B1, Sum Aflatoxin B1+B2+G1+G2 in cereals, nuts and dried products - HPLC in the range of 1-10 µg/kg for G1 and B1, 0.3-2.5 µg/kg for G2 and B2

What was the extraction solvent used?
What was the extraction mode (e.g. blending, shaking)?
What was the extraction time?
What was the solvent to sample ratio used during extraction (in mL/g)?
Were any extraction aids added? If yes, please specify

Lab Code	Extraction solvent	Extraction mode	Extraction time [min]	Solvent to sample ratio volume [ml]	Extraction aids added (e.g. NaCl)?
401	MeOH/H2O 4:1	High speed blending + shaking + high speed blending	2 min + 30 min + 2 min	5:1	No
403	60% acetonitrile, 40% water	Turrex blender	2 minutes	4	No
404	84:16 acetonitrile:water	Blending	4 minutes	8	No
405	methanol/water (80/20; v/v)	3 min ultra-turrax, then 30 min stiring	3 min ultra-turrax, then 30 min stiring	10	2.5 g NaCl
406	Acetonitrile:water 80:20	shaking	20 mins	8ml/2g	No
407	MeOH:H2O (4:1)	Blending	3 minutes, high speed	80mL/20g (4mL/g)	5g
408	acetone/isopropanol/water/acetic acid (15/5/14.9/0.1)	sonication (10 minutes) followed by overhead mixing (60 minutes)	70 minutes total (excluding the QuEChERS step)	35 ml solvent to 4 grams sample	QuEChERS based extraction (8g MgSO4/1.5g NaCl, added after completion of the extraction at 70 min), no d-SPE clean-up was used
409					
410	methanol	shaking	20 min.	3.5	No
411	Methanol/ Water 70/30	Blending	2 min	10g /100 ml	5g NaCl
412	Methanol 70%	Mixing.	30 min	Methanol 70%, 5 ml/g	No
413	MeOH:H2O (70:30)	shaking	30 min	in 20 mL/ 5g	No
414	extraction solvent (extraction solvent is 70 : 30 Methanol : water)	Blending	2 min	125 ml of extraction solvent (extraction solvent is 70 : 30 Methanol : water)	a blender jar weigh 25 g of sample then add 5 g of sodium chloride
415	Methanol:Water (60:40)	Blending	1 min	10	NaCl, 5g
416	methanol	sonication	15 minutes	5 mL/1g	0.4 g
417	Methanol-water	shaking	1 hour	62,5 mL / 6.25 g	0.5 g
418					
419	MeOH/H2O	Blending	2min	125 mL/25 g	NaCl
420	MeOH/H2O	shaking	30 minutes	25 ml / 6.25 g	No
421					
422	acetonitile	homogenization	1 min	5 : 1	No
423	80% methanol/ water	Blending	3 min.	20 ml/ 5 g	NaCl 0.5 g
424	Methanol 70%	Blending	15 min.	25/5	No
425					
426					
427	Methanol:Water 80:20	Blending	3 minutes	150mL solvent/25g sample	NaCl
428					
429	Methanol : water = 4:1	Blending by polytron.	3 minutes	5mL/g	No
430	MeOH/H2O: 80/20	Blending	2 minutes	200ml/13l	5 g NaCl
431					
432	methanol-water (80-20)	Blending	3 min	4 ml/g	NaCl 2,5 g / 25 g
433	methanol	shaking	1,5 hours	3/1	0,08g Na Cl /1 g sample
434					
435	methanol	Blending	30 minutes	5/1	0.25 gNaCl for 5g sample
436	80% Methanol	High-speed blending	3 minutes	4 ml/g	NaCl; 2.5 g
437	MeOH-Water	Blending	3 min	4	2,5 g
438	Methanol:water	shaking	30 minutes	25 ml/5 g	NaCl
439	ACN: 0,1% HCOOH in H2O (1:1); modified QuEChERS	Blending	20 min	5 mL/g	1 g of NaCl; 4 g of MgSO4
440	acetonitril:water (v/v 70:30)	mixing	2 h	50/5	No
441	Methanol - water (80:20 v/v)	Blending	3 minutes	4	2.5 g before blending
442					
443	80%MeOH	Blending	15min	10g/100ml	No
444	acetone:water 85:15	shaking	30min	250/50	No
445					
446					
447	Methanol/Water (8/2)	Ultra-Turrax	3 min.	5 mL/g	NaCl, 5g
448	Methanol/Water 62.5 % in methanol	Blending with Ultra-Turrax	20 min	4.0 mL/g	NaCl
449	Methanol 70%	Shaker	15 minut	5 g sample with 25 ml of 70%	1 ml distilled water

Lab Code	Extraction solvent	Extraction mode	Extraction time [min]	Solvent to sample ratio volume [ml]	Extraction aids added (e.g. NaCl)?
				methanol	
450	Methanol: water (70:30)	Blending	2 minutes	5 mL/g	5 g per 25 g of sampple
451	acetone/water 85:15 v:v	turbulent shaking overhead	40 min	6 mL/g	1 g NaCl to 30 mL extraction solvent
452	60 %Acetonitirle and 40 % Water	Blending	2 minutes	3.76	No
454	Acetonitrile : water	shaking	2 hours	70:30	No
455	Acetonitrile:water, 60:40, v/v	Blending	4 minutes	100mL/10g	No
456	Acetone/Water	Blending/shaking	30 minutes	10 g to 50 ml	No
457	80 % methanol	shaking	3 hours	4: 1	NaCl, 5 g, 100 ml n-hexan
458	methanol/water	sonication	10 min	3	No
459	methanol/water	Blending	90 minutes	10	1.6 g NaCl on 20 g sample
461	MeOH + H2O	shaking	3 min	7	hexane 30ml, NaCl 5g

What type of clean up methodology was used (e.g. immunoaffinity column)? Please specify manufacturer, brand and the production code.
Specify the detection method used.
Which derivatisation method was applied?
How did you integrate the signals? Was automatic integration confirmed for correctness visually?
Did you use acid washed glassware? Was protection against daylight applied?

Lab Code	Clean-up	Manufacturer of IAC	Brand and production code of IAC	Determination technique	Specification of the method used	Derivatisation agent	Peak intergation type	Visual check?	Acid washed glassware?	Protection against daylight
401	Immunoaffinity column	Vicam	Aflatest WB	HPLC-FLD	GraceSmart RP18, 5 µm, 150×4.6 mm, MeOH/AcCN/H2O 30:20:70 + KBr + HNO3	Kobra cell	Automatic	Yes	Yes	Yes
403	Immunoaffinity column	Rhone Biopharm	Easi - Extract aflatoxin affinity columns	HPLC-FLD	ODSI 5 micron, 25cm	Kobra cell	Automatic	Yes	No	No
404	MultiSep AflaZON 226			HPLC-FLD	Column Ace3 C18 100x4.6 mm, mobile phase water:acetonitrile:methanol 900:180:240 + KBr+ HNO3	Kobra cell	Automatic	Yes	Yes	Yes
405	IAC	r-biopharm	Aflaprep; BC117	HPLC-FLD	Zorbax Eclipse XDB-C18; 5µ; 250*4.6 mm; ACN/methanol/acetic acid 2% (18/27/55; v/v/v); flow=1ml/min	PBPB	Manual	true	No	Yes
406	none			LC-MSMS		NA	Automatic	Yes	No	Yes
407	IAC	R-Biopharm - Easi-extract	BB994/50	HPLC-FLD	Symmetri C18 (5micron - 4.6x250mm); mobile phase 54:29:17 (H2O:MeOH:AcCN); 1mL/min; post column derivatisation 0,4 mL/min	PBPB	Automatic	Yes	Yes	Yes
408	No additional clean-up			LC-MS/MS	Phenomenex XB-C18 column, M+H ion, 1mg equivalent injected	none	Automatic	Yes	Yes	Yes
409										
410	-	-	-	photometrically at 450 nm	-	-	true	true	No	Yes
411	Immunoaffinity column	Vicam- USA	Vicam AflaTest, Reorder #G1010, lot 1911E	HPLC-FLD	Zorbax eclipse C18, 250mm, 4,6µm, 5µ; Acetonitrile:methanol:water(KBr, HNO3)	Kobra cell	Manual	true	Yes	Yes
412	Filtration.			ELISA		Not applicable.	Automatic	Yes	No	Yes
413	Immunoaffinity column	r-biopharm	R-Biopharm, RP70N	HPLC-FLD	ODS HYPERSIL 250x4.6 mm (5 um), 600:200:200 (H2O:ACN:MeOH)+ 100 ul HNO3, 0,119g KBr	Kobra cell	Automatic	Yes	No	Yes
414	afla star	biopharm	aflapred	HPLC-FLD	HPLC Column/Water, high purity650+ Acetonitrile, HPLC grade 175 ml + Methanol, HPLC grade 175 ml		Automatic	Yes	No	No
415	Immunoaffinity column	Neogen	Neocolumn for Aflatoxins-Wide Bore, Code 8043	HPLC-FLD	C18, Water:Methanol:Acetonitrile (64:23:13)	Electrochemical derivatization	Manual	true	Yes	Yes
416	Immunoaffinity column	r-biopharm	R-Biopharm easi-extract aflatoxin RP70N	HPLC-FLD	column C18, mobile phase-methanol:acetonitril: water 3:2:6 v/v/v +120 mg KBr + 350 µl HNO3	Kobra cell	Manual	true	No	Yes
417	Imunoaffinity column	R-Biopharm Rhone	Aflaprep, P07	HPLC-FLD	Prodigy C18(2), 100A, 250x4.6 mm (Phenomenex), acetonitrile-methanol-water, addition of KBr, HNO3	Kobra cell	Automatic	Yes	No	Yes
418										

Lab Code	Clean-up	Manufacturer of IAC	Brand and production code of IAC	Determination technique	Specification of the method used	Derivatisation agent	Peak intergation type	Visual check?	Acid washed glassware?	Protection against daylight
419	Immunoaffinity column	r-biopharm	Aflaprep P07	HPLC-FLD	Lichrospher 100 RP18 10 um 250mm, 4mm; MeOH/ACN/H2O; Flow:1,2mL/min; Ex:350 nm, Em:435 nm	Iodine	Manual	true	No	Yes
420	Immunoaffinity column	Romer Labs	AflaStar R	HPLC-FLD	Luna 5u C18 (150x4.6), 100 A; H2O/MeOH/KBr/4M HNO3; flow 0.9 ml/min; time of analysis 20 minutes	Kobra cell	Manual	true	No	Yes
421										
422	Immunoaffinity column	Vicam	G1024	HPLC-FLD	C8 Inertsil, acetonitrile:methanol:H2O (8:27:65)	TFA	Automatic	Yes	No	Yes
423	Immunoaffinity column	r-biopharm	RP70N EE AFLA 50 Bx: AG 831/50	HPLC-FLD	LiChroCART 250-4 LiChrospher 100 RP-18 endcapped (5 µm); KBr,HNO3 solution/ACN/MeOH (6/2/3)	Kobra cell	Manual	Yes	Yes	Yes
424	Immunoaffinity column	Ridacreen	R-Biopharm	ELISA		no	true	true	No	Yes
425										
426										
427	Immunoaffinity column	Vicam	AFLATEST Lot No. 1957E	HPLC-FLD	Column, C18 Kinetix 2.6u PFP 100Å; Mobile phase, Methanol:Water 45:55.	Photochemical Reactor for Enhanced Detection "PHRED"	Automatic	Yes	Yes	Yes
428										
429	immunoaffinity columns	LCTech in Germany	P/N 11022	HPLC-FLD	Phenomenex Luna 3um C18 (2); water with HNO3 and KBr/methanol/acetonitril	CoBrA-cell from Coring - System	Automatic	Yes	No	Yes
430	IAC	AFLAPREP, Rhone Diag.	P07	HPLC-FLD	Spherisorb 5 ODS-1, 250x4,6mm, H2O/ACN/MeOH: 57/20/23	PBPB	Automatic	Yes	Yes	Yes
431										
432	Immunoaffinity column	R-Biopharm Rhone Ltd	EASI-EXTRACT AFLATOXIN, Code RP70N, Batch BB981, ExpDate 10August2015	HPLC-FLD	RP-C18, 3,9 x 300 mm, 4 um, MobPhase Acetonitrile-Methanol-Water (140-290-570)	PBPB (Pyridinium hydrobromide perbromide)	Automatic	Yes	Yes	Yes
433	Immunoaffinity column	r-biopharm	aflaprep	HPLC-FLD	C18 250 x 4,6 mm; wather-methanol-acetonitrile:57/38/5	Kobra cell	Automatic	Yes	No	Yes
434										
435	Immunoaffinity column	easi- extract aflatoxin - R Biopharm	RP70N	HPLC-FLD	Column Eclipse PlusC18, mobile phase - methanol 40%, KBr , HNO3 4M	Kobra cell	Automatic	Yes	No	Yes
436	Immunoaffinity column	R-Biopharm Rhone Ltd	r-Biopharm, AFLAPREP P07	HPLC-FLD	Column: Supelcosil 25 cm x 4.6 mm x 5 um; Mobile Phase: Water / Methanol / Acetonitrile (60/30/20)	Bromination (PBPB)	Automatic	Yes	No	Yes
437	IAC	Neogen	Neocolumn Production code : 8043	HPLC-FLD	LichroCart RP18 5 µm 125 x 4 mm MP : MeOH-ACN- H2O	Kobra cell	Automatic	Yes	No	No
438	Immunoaffinity column	R-Biopharm Rhone Ltd	Aflaprep P07	HPLC-FLD	Waters Spherisorb 3µm ODS2 150mm x 4.6mm, water:methanol:acetonitrile:tetrahydrofuran, Ex 340nm Em 430nm	Post column derivatization PBPB in water	Manual	true	No	Yes
439	QuEChERS, dilution with water			LC-MS	Column: ACQUITY UPLC BEH C18, 50 mm x 2.1 mm x 1.7 microm; Gradient elution;	no derivatisation	Automatic	Yes	No	Yes

Lab Code	Clean-up	Manufacturer of IAC	Brand and production code of IAC	Determination technique	Specification of the method used	Derivatisation agent	Peak intergation type	Visual check?	Acid washed glassware?	Protection against daylight
					Mobile phase A: 0.1 % HCOOH in water, Mobile phase B: 1mM HCOONH4 + 0.1 % HCOOH in MeOH; Ions: 313.1 > 241.1, 313.1 > 213, mass fraction of sample injected: 0.25 mg.					
440	Immunoaffinity column	Vicam	Vicam, AflaTest WB	LC-MS	Column C18; mobile phase 0.1% formic acid in water and 0.1% formic acid in acetonitril; SRM transitions: 313-241 and 313-285; injected 40 mg of sample.	None.	Automatic	Yes	Yes	No
441	immunoaffinity columns	R-Biopharm Ltd	EASI-EXTRACT AFLATOXIN RP70N	HPLC-FLD	Inertsil ODS-2, 5um, 150 mmx4.6 mm; water- acetonitrile-methanol (60:10:30 v/v) + Kobra cell 119 mg KBr and 100 ul HN03 in 1 litre mobile phase; flow rate 1.0 ml/min; FLD Ex=362 nm, Em=435 nm; injection volume 100 ul	Kobra cell	Manual	true	Yes	Yes
442										
443	Immunoaffinity column Easi extract aflatoxin; Rhone biopharm	Rhone Biopharm	Easi extract aflatoxin;	LC-MS	phenomenex PFP; 3um, 2.1×150mm, 313, 30ul	no derivatisation	Manual	true	No	Yes
444	Immunoaffinity column	LCTech GmbH	AflaCLEAN Select, production code: 12058	HPLC-FLD	Symmetry C18 (4.6x250mm), water:methanol:acetonitrile 50:40:10	photochemical derivatosation	Automatic	Yes	No	Yes
445										
446										
447	Immunoaffinity column	r-biopharm	Aflaprep, Code P07	HPLC-FLD	Column: Lichrospher RP-18 (250x4.6mm, 5um), Mobile Phase: Water/Methanol/Acetonitril (56/22/22) + 119 mg/L KBr + 350 uL/L HNO3 (4 mol/L)	Kobra cell	Automatic	Yes	Yes	Yes
448	Immunoaffinity column	Neogen	Brand: NeoColumn, Production Code: 8043	HPLC-FLD	Type of Column: Waters Symmetry 4.6x250mm/5µm/ODC(C18), Mobile Phase: Tetrahydrofuran/Water 21% in Tetrahydrofuran, Injection Volume 200 µL	Saturated Iodine	Manual	true	No	Yes
449	I'm not use.			Elisa method		I'm not use.	Manual	true	No	Yes
450	Immunoaffinity column	Vicam	Aflatest Lot 1881	HPLC-FLD	Zorbex Eclips C18, (ACN:Water:MeOH) (175:650:175 ml)	Post column derivatization with pyridinium hydrobromide perbromide	Automatic	Yes	No	Yes
451	IAC	Vicam	WB 1942 N	HPLC-FLD	Gemini C18 250 x 4.6 mm, 5 µm; water/methanol/acetonitrile 54/29/17 v:v:v + 119 mg/L KBr + 100 µL/L HNO3	Kobra cell	Automatic	Yes	No	Yes
452	Immunoaffinity column	rhone ltd	aflaprep	HPLC-FLD	10um C18 ODS column. eluent water + acetonitirle + methanol mix. detection wavelenghts Ex 365 nm Em 455nm	50ul trifluoroacetic acid	Automatic	Yes	Yes	No
454	none			LC-MS	ACQUITY BEH UPLC C18, 2.1 X 100 mm, 1.7 µm	none	Automatic	Yes	No	Yes
455	Immunoaffinity column	R-Biopharm Rhone	Aflaprep P07	HPLC-FLD	Spherisorb ODS1, 25cm x 4.6mm id, Acetonitrile:water:methanol. 56:30:14, v/v/v	Kobra cell	Automatic	Yes	Yes	Yes

Lab Code	Clean-up	Manufacturer of IAC	Brand and production code of IAC	Determination technique	Specification of the method used	Derivatisation agent	Peak intergation type	Visual check?	Acid washed glassware?	Protection against daylight
456	immunoaffinity columns	Aflaprep R_Biopharm Rhone	P07, batch AC760	HPLC-FLD	Waters Nova Pak C-18, 4 µm, 3.9 x 150 mm	Jodine	Automatic	Yes	No	No
457	immunoaffinuty column	R-Biopharm Rhone	Easi-extract aflatoxin, BX:AB 728/50, EEAfla 50	HPLC-FLD	Nucleosil 100-5C8 25/4.6; mobile phase 600 ml water:200 ml ACN:300MeOH, 120mgKBr, 350 microl HNO3 4M	Kobra cell	Manual	true	No	Yes
458	Immunoaffinity column	Rommer	Aflastar	HPLC-FLD	C-18, KBr/HNO3/acetonitrile/H20	Kobra cell	Automatic	Yes	Yes	Yes
459	Immunoaffinity column	R-Biopharm Rhone Ltd	AFLAPREP, BX: BB983/50, product code: P07	HPLC-FLD	Column: Agilent Eclipse XDB-C18 150 x 4.6 mm, 5 um; MP: Acetonitrile/methanol/water; post-column derivatisation, 60 °C	Iodine	Manual	true	Yes	Yes
461	IA column	R-Biopharm Rhone	EASI-EXTRACT, AFLATOXIN, Lot: BC105150	HPLC-FLD	Luna 100A C18, 250 x 4.6 mm, 5 µm, mob. phase H20, KBr, CH3CN, 4M HN03, MeOH		Manual	true	Yes	Yes

Did you encounter any problems during the analysis?
Did you need to include any overnight stop during the analysis?
Did you notice any unusual observations which, however, did not seem to have any effect on the results?

Lab Code	Problems	Overnight stops	Unusual observations
401	Yes, The material had a trend to aggregate during extraction.	No	Yes, The material had a trend to aggregate during extraction.
403	No	No	No
404	No	Yes	No
405	No	No	No
406	Yes, Clogging of samples level A and B	No	Yes, there seemed to be a lot of moisture in samples, especially level A and B. Samples were very clogged.
407	No	No	No
408	Yes, we doubled the volume of extraction solvent compared to our routine method, as the samples were too greasy	No	No
409			
410	No	No	No
411	No	No	No
412	No	No	No
413	No	No	No
414	No	No	No
415	Yes, We didn't use the Blank Sample to calculate Recovery.	No	Yes, Samples A and B were just enough for our Method and not well homogenised
416	No	No	No
417	No	No	No
418			
419	No	No	No
420	Yes, During the extraction a solid precipitate of vegetable fat was formed	No	No
421			
422	No	No	No
423	No	Yes	No
424	No	No	No
425			
426			
427	No	Yes	No
428			
429	No	Yes	No
430	No	Yes	No
431			
432	Yes, The material of blank sample used for recovery test was not of same type as material of samples A and B. Despite of that the recovery received was used for calculations.	No	No
433	No	Yes	Yes, After extraction with methanol and centrifugation has developed a greasy film and after clean-up the final elute was milky.
434			
435	No	No	No
436	No	No	No
437	No	No	Yes, Interference near AF B2 and AF G1 appears after 24H. When diluting extract we observe a decrease of AF B1 Signal
438	No	No	No
439	No	No	No
440	No	No	No
441	No	No	No
442			
443	No	No	No
444	No	No	No
445			
446			
447	No	No	No
448	No	No	No
449	No	No	No
450	No	No	No
451	Yes, not enough sample material to obtain representative sample weights according to the standardized method used; homogeneous weighted portions were difficult to made because of the material consistency	No	No
452	No	No	No
454	No	No	No

Lab Code	Problems	Overnight stops	Unusual observations
455	No	No	No
456	Yes, Clouded sample was resolved with warm water	Yes	No
457	No	No	No
458	No	No	No
459	No	No	No
461	No	No	No

Did you find the instructions distributed for this PT adequate?
Any Other comments you wish to make?

Lab Code	Instructions adequate?	Registering format	Any Other comments
401	Yes	Slightly onerous – but of understandable reasons.	-
403	Yes		
404	Yes	It is not clear how to go back to the main submitting site from the questionnare	
405	Yes	good	
406	Yes	good	no
407	Yes	We find it usefull and appropriate	-
408	Yes	good	no
409			
410	Yes	Very good	no
411	Yes	Very good	no
412	Yes	Very useful.	No, Thank you.
413	Yes	It was appropriate.	
414	Yes	very well	thank tou
415	Yes		
416	Yes	the interface is easy to use	
417	Yes	Appropriate	
418			
419	Yes	It's good	
420	Yes	Normal	
421			
422	Yes	O.K.	
423	Yes		
424	Yes	registering / reporting format is complete	no
425			
426			
427	Yes	Well organised and easy to follow	We normally take 50g of sample for analysis but as only 25g was supplied per sample we had to adjust our method accordingly.
428			
429	Yes	-	-
430	Yes	fine	
431			
432	Yes	Okay	It would be nice if the amount of the sample were 50 g.
433	Yes	Will be better if to report the results and to fill in the questionnaire to be on only one log in; because at this moment are necessary to make two log in (one for questionnaire, and another for results)	no
434			
435	Yes	no	I want to participate at mycotoxins worshop as official laboratories for multi-residue methods
436	Yes	Satisfactory	
437	Yes	It is OK	Next time could we have more Blank sample to ananlyse without spiking
438	Yes	This report format was rather general. We didn´t know how detailed method information was wanted. That´s why we didn´t mention for example volume ratios, dilution with water and filtering the sample extraction liquid, etc.	
439	Yes		
440	Yes		
441	Yes	OK!	
442			
443	Yes	OK	no
444	Yes	Very good	no
445			

Lab Code	Instructions adequate?	Registering format	Any Other comments
446			
447	Yes	Satisfactory	
448	Yes		
449	Yes	It's OK	No, but I'm expected for payment formalities! Thanks for all!
450	Yes		
451	Yes		General comment: for future PT it would be useful to increase the sample quantities; Measurement uncertainty could not be determined because of the small sample quantities, three sample weights of min. 10 g each would have been necessary to calculate the MU
452	Yes		We need a min of 50g of sample for analysis next time could more of the sample be issued
454	Yes	It's OK	The amount of this P.T was not enough
455	Yes		
456	No, The instruction about clouding sample came a little too late	Reporting is complicated and for detailed. It is the 1. time for me. Why not use input online, 1 for the results and 1 for the method, like all other providers of PT do. So that I not have to print out and scan and send by mail. The pop up window about saving your questionnaire drives me crazy!	The specified recoveries are 2: 92 % measured for this analysis and 88% as fixed used for the results. May be you should distinguish between those possibilities
457	Yes	good	-
458	Yes	is correct, no problem	no
459	Yes	Everything was properly	None
461	Yes		

European Commission
EUR 26849 EN – Joint Research Centre – Institute for Reference Materials and Measurements

Title: **Report on the 2014 Proficiency Test of the European Union Reference Laboratory for Mycotoxins. Determination of Aflatoxin B1 in Copra (Coconut powder)**

Author(s): Maciej Kujawski, Carsten Mischke, Joerg Stroka

Luxembourg: Publications Office of the European Union

2014 – 38 pp. – 21.0 x 29.7 cm

EUR – Scientific and Technical Research series – ISSN 1831-9424 (online)

ISBN 978-92-79-41181-6 (PDF)

LA-NA-26849-EN-N

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doi:10.2787/98787

ISBN 978-92-79-41181-6

Publications Office