Evaluation Report

proficiency test

DLA 12/2019

Allergen-Screening II:

Crustaceae, Egg, Fish, Milk, Molluscs, Mustard and Soybean

DLA - Proficiency Tests GmbH
Kalte Weide 21
24641 Sievershütten/Germany

proficiency-testing@dla-lvu.de *www.dla-lvu.de*

Coordinator of this PT:
Matthias Besler-Scharf, Ph.D.

Allgemeine Informationen zur Eignungsprüfung (EP)
General Information on the proficiency test (PT)

EP-Anbieter *PT-Provider*	**DLA - Proficiency Tests GmbH** Kalte Weide 21, 24641 Sievershütten, Germany Geschäftsführer/CEO: Dr. Matthias Besler-Scharf Stellv. Leitung/Deputy Lead: Alexandra Scharf MSc. Tel. ++49-(0)4532-9183358 Mob. ++49(0)171-1954375 Fax. ++49(0)4102-9944976 eMail. proficiency-testing@dla-lvu.de
EP-Nummer *PT-Number*	DLA 12/2019
EP-Koordinator *PT-Coordinator*	Dr. Matthias Besler-Scharf
Status des EP-Bericht *Status of PT-Report*	Abschlussbericht / Final report (15 November 2019) Gültig ist die jeweils letzte Version/Korrektur des Berichts. Sie ersetzt alle vorangegangenen Versionen. Only the latest version/correction of the report is valid. It replaces all preceding versions.
EP-Bericht Freigabe *PT-Report Authorization*	Dr. Matthias Besler-Scharf (Technischer Leiter / Technical Manager) *- gezeichnet / signed M. Besler-Scharf* Alexandra Scharf MSc. (QM-Beauftragte / Quality Manager) *- gezeichnet / signed A. Scharf* Datum / Date: 15 November 2019
Unteraufträge *Subcontractors*	Falls im Rahmen der Eignungsprüfung eine Prüfung der Gehalte, Homogenität und Stabilität von EP-Parametern durchgeführt wurde, hat DLA diese im Unterauftrag vergeben. In case the analysis of the content, homogeneity and stability of PT-parameters was part of the proficiency test, the determinations were subcontracted by DLA.
Vertraulichkeit *Confidentiality*	Die Teilnehmerergebnisse sind im EP-Bericht in anonymisierter Form mit Auswertenummern benannt. Daten einzelner Teilnehmer werden ausschließlich nach vorheriger Zustimmung des Teilnehmers an Dritte weitergegeben. Participant result are named anonymously with evaluation numbers in the PT report. Data of individual participants will be passed on to third parties only with prior consent of the participant.

Inhalt / Contents

1. Introduction

The participation in proficiency testing schemes is an essential element of the quality-management-system of every laboratory testing food and feed, cosmetics and food contact materials. The implementation of proficiency tests enables the participating laboratories to prove their own analytical competence under realistic conditions. At the same time they receive valuable data regarding the verification and/or validation of the particular testing method [1, 5].
The purpose of DLA is to offer proficiency tests for selected parameters in concentrations with practical relevance.
Realisation and evaluation of the present proficiency test follows the technical requirements of DIN EN ISO/IEC 17043 (2010) and DIN ISO 13528:2009 / ISO 13528:2015 [2, 3].

2. Realisation

2.1 Test material

Four PT-samples were provided for the qualitative detection of allergens in mg/kg range. To prepare the samples premixes were used at levels of about 2,4-10% of the allergenic ingredients concerned.
The respective raw materials for the allergens used were commercial egg powder, milk powder and soy flour and premixes produced by DLA from commercial mustard seeds as well as frozen crayfish (cooked, peeled), salmon and squid (s. Tab. 2). The mustard seeds were crushed, ground with addition of carrier substances and sieved (mesh 400 µm). The frozen products were crushed, dried and ground with addition of carrier substances and sieved by means of a centrifugal mill (mesh 250 µm).

The composition of the allergen-premixes is given in table 1. The premixes were used for spiking of the PT-samples 1 to 4 (see Tab. 2).

After homogenisation the samples were portioned to approximately 20 g into metallised PET film bags.

Table 1: Composition of DLA-Samples

Ingredients	**Samples 1 - 4**
Potato powder (Ingredients: Potatoes, E471, E304, E223, E100)	74 - 76 %
Maltodextrin	24 - 26 %
Allergen-Premixes Ingredients: - Maltodextrin (30% - 88%) - Titanium dioxide (0,0% - 40%) - Sodium sulfate (0,0% - 7,7%) - Silicon dioxide (1,0% - 2,2%) - Allergens (2,4% - 10% each)	0,2 - 0,8 %

Table 2: Added amounts of allergenic ingredients positive in mg/kg ranges** given as food item

Ingredients *	Sample 1	Sample 2	Sample 3	Sample 4
Crustaceae: Louisiana Crayfish *(Procambarus clarkii)*, dried (Protein 79%)	negative	positive (75 - 150)	positive (25 - 75)	negative
Egg: Whole egg powder (Protein 47%)	positive (50 - 100)	negative	positive (75 - 150)	negative
Fish: Salmon *(Salmo salar)*, dried (Protein 54%)	negative	positive (25 - 75)	negative	positive (50 - 150)
Milk: Skimmed milk powder (Protein 37%)	positive (50 - 150)	negative	negative	positive (25 - 75)
Molluscs: Squid tubes *(Illex argentinus)*, dried (Protein 34%)	positive (25 - 75)	negative	negative	positive (75 - 150)
Mustard, yellow: Sinapis alba (Protein 31%)	negative	positive (50 - 150)	negative	negative
Mustard, brown: Brassica juncea (Protein 28%)	negative	negative	positive (50 - 150)	negative
Mustard, black: Brassica nigra (Protein 27%)	positive (50 - 150)	negative	negative	negative
Soya: Soyflour, not toasted (Protein 37%)	negative	positive (50 - 150)	positive (25 - 75)	negative

* Protein contents according to laboratory analysis (total nitrogen, Kjeldahl general factor F=6,25)
**Allergen contents of „food item" as indicated in the column of ingredients according gravimetric mixing
Note: *The metrological traceability of temperature, mass and volume during production of the PT samples is ensured by DAkkS calibrated reference materials.*

The detectability or absence of the allergens was tested by DLA using lateral flow assays. The results are in agreement with the spiking of the PT samples 1-4 (see Table 3).

Table 3: Verification of detectability of the added allergens by lateral flow assays (AgraStrip® LFD, Romer Labs®)

Lateral Flow Device (LFD)*	Sample 1	Sample 2	Sample 3	Sample 4
AgraStrip® Crustaceae	negative	positive	positive	negative
AgraStrip® Egg	positive	negative	positive	negative
AgraStrip® Casein	positive	negative	negative	positive
AgraStrip® Soy	negative	positive	positive	negative
AgraStrip® Mustard	positive	positive	positive	negative

* Nachweisgrenze jeweils 2-10 mg/kg / Limit of detection (LOD) 2-10 mg/kg each

2.1.1 Homogeneity

The **mixture homogeneity before bottling** was examined 8-fold by **microtracer analysis.** It is a standardized method that is part of the international GMP certification system for feed [14].
Before mixing dye coated iron particles of µm size are added to the sample and the number of particles is determined after homogenization in taken aliquots. The evaluation of the mixture homogeneity is based on the Poisson distribution using the chi-square test. A probability of ≥ 5 % is equivalent to a good homogeneous mixture and of ≥ 25% to an excellent mixture [14, 15].
The microtracer analysis of the present PT samples 1-4 showed probabilities of 57%, 96%, 92% and 56%, respectively. Additionally particle number results were converted into concentrations, statistically evaluated according to normal distribution and compared to the standard deviation according to Horwitz. For the assessment HorRat values between 0,3 and 1,3 are to be accepted under repeat conditions (measurements within the laboratory) [17]. This gave HorRat values of 1,1, 0,5, 0,7 bzw. 1,1, respectively. The results of microtracer analysis are given in the documentation.

2.1.2 Stability

A water activity (a_W) of < 0,5 is an important factor to ensure the stability of dry or dried products during storage. Optimum conditions for storage is the a_W value range of 0,15 - 0,3. In this range the lowest possible degradation rate is to be expected [16].

The experience with various DLA test materials showed good storage stability with respect to the durability of the sample (spoilage) and the content of the PT parameters for comparable food matrices and water activity (a_W value <0,5).
The a_W value of the PT samples was approx. 0,27 - 0,30 (20-22°C). The stability of the sample material was thus ensured during the investigation period under the specified storage conditions.

2.2 Sample shipment and information to the test

The portions of the test materials (sample 1 to 4) were sent to every participating laboratory in the 27th week of 2019. The testing method was optional. The tests should be finished at August 30th 2019 the latest.

With the cover letter along with the sample shipment the following information was given to participants:

There are 4 different samples possibly containing the allergenic ingredients Crustaceae, Egg, Fish, Milk, Molluscs, Mustard (yellow/white, brown and black) and/or Soybean in a simple carrier matrix The evaluation of results is strictly qualitative (positive / negative).

The following analysis methods can be used:

a) ELISA and Lateral Flow
b) PCR

Please note the attached information on the proficiency test.
(see documentation, section 5.3 Information on the PT)

2.3 Submission of results

The participants submitted their results in standard forms, which have been sent by email. The results given as positive/negative were evaluated.
Queried and documented were the indicated results and details of the test methods like specificities, test kit manufacturer and hints about the procedure.
In case participants submitted several results for the same parameter obtained by different methods these results were evaluated with the same evaluation number with a letter as a suffix and indication of the related method.

Out of 35 participants 34 submitted at least one result in time. One participant submitted no results.

3. Evaluation

Different ELISA- and PCR-methods for the determination of allergens in foods are eventually using different antibodies and target-DNA, are usually calibrated with different reference materials and may utilize differing extraction methods. Among others this can induce different valuation of the presence and/or content of the analyte [25, 26, 27, 28]. Furthermore matrix- and/or processing of samples can have strong impact on the detectability of allergens by ELISA and PCR methods.

Therefore in the present PT the allergenic ingredients were provided for analysis in a simple matrix without further processing.

3.1 Agreement with consensus values from participants

The qualitative evaluation of the ELISA and PCR results of each participant was based on the agreement of the indicated results (positive or negative) with the **consensus values from participants**. A consensus value is determined unless ≥ 75% positive or negative results are present for a parameter.
The assessment will be in the form that the number of matching results followed by the number of samples for which a consensus value was obtained is indicated. Behind that the agreement is expressed as the percentage in parentheses.

3.2 Agreement with spiking of samples

The qualitative evaluation of the ELISA and PCR results of each participant was based on the agreement of the indicated results (positive or negative) with the **spiking of the four PT-samples**.
The assessment will be in the form that the number of matching results followed by the number of samples is indicated. Behind that the agreement is expressed as the percentage in parentheses.

4. Results

All following tables are anonymized. With the delivering of the evaluation-report the participants are informed about their individual evaluation-number.

The qualitative evaluation is carried out for each parameter for ELISA and PCR methods separately. Results of lateral flow methods were valuated together with ELISA methods, because they are usually based on antibody detection.

The participant results and evaluation are tabulated as follows:

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive				
Number negative				
Percent positive				
Percent negative				
Consensus value				
Spiking				

4.1 Proficiency Test Crustaceae

4.1.1 ELISA-Results: Crustaceae (Crayfish)

Qualitative valuation of results

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
1	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	AQ	
2	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	AQ	
17	positive	positive	negative	negative	2/4 (50%)	2/4 (50%)	AQ	
26	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	AQ	
24	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	BF	
34	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	BF	
5	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	EF	
21	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	EF	
28	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	EF	
30	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	EF	
14	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	ES	
23	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	IL	
7	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	NL-E	
3	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
6	positive	positive	positive	positive	2/4 (50%)	2/4 (50%)	RS-F	
12	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
20	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
32	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
13	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	VT	

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	2	19	18	1
Number negative	17	0	1	18
Percent positive	11	100	95	5
Percent negative	89	0	5	95
Consensus value	negative	positive	positive	negative
Spiking	negative	positive	positive	negative

Methods:

AQ = AgraQuant, RomerLabs

BF = MonoTrace ELISA, BioFront Technologies

EF = SensiSpec ELISA Kit, Eurofins

ES = ELISA-Systems

IL = Immunolab

NL-E = nutriLinia®E Allergen-ELISA

RS = Ridascreen®, R-Biopharm

VT = Veratox, Neogen

Comments:

The consensus values of results are in qualitative agreement with the spiking of samples.

4.1.2 PCR-Results: Crustaceae (Crayfish)

Qualitative valuation of results

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
3	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	SFA	
7	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	SFA	
14	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	SFA	
24	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	SFA	
28	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	SFA	
29	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	SFA	
33	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	SFA	

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	0	7	7	0
Number negative	7	0	0	7
Percent positive	0	100	100	0
Percent negative	100	0	0	100
Consensus value	negative	positive	positive	negative
Spiking	negative	positive	positive	negative

Methods:

SFA = Sure Food ALLERGEN, R-Biopharm / Congen

Comments:

The consensus values of results are in qualitative agreement with the spiking of samples.

4.2 Proficiency Test Egg

4.2.1 ELISA-Results: Egg (Whole egg powder)

Qualitative valuation of results

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
2	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	AQ	
4	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	AQ	
10	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	BC	
34	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	BF	
6	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	BK	
5	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	EF	
28	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	EF	
18	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	ES	
23	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	IL	
1	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	MI	
26	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	MI	
16	positive	positive	positive	positive	2/4 (50%)	2/4 (50%)	MI-II	
21	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	MI-II	
24	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	MI-II	
25	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	RS	
7a	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
9	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
12	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
14	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
17	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
19	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
20	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
32	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
7b	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	RS-L	Lysozyme

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	24	1	24	1
Number negative	0	23	0	23
Percent positive	100	4	100	4
Percent negative	0	96	0	96
Consensus value	positive	negative	positive	negative
Spiking	positive	negative	positive	negative

Methods:

AQ = AgraQuant, RomerLabs
BC = BioCheck ELISA
BF = MonoTrace ELISA, BioFront Technologies
BK = BioKits, Neogen
EF = SensiSpec ELISA Kit, Eurofins
ES = ELISA-Systems
IL = Immunolab
MI = Morinaga Institute ELISA
MI-II = Morinaga Institute ELISA II
RS = Ridascreen®, R-Biopharm
RS-F= Ridascreen® Fast, R-Biopharm
RS-L= Ridascreen® Lysozyme, R-Biopharm

Comments:
The consensus values of results are in qualitative agreement with the spiking of samples.

4.2.2 PCR-Results: Egg (whole egg powder)

Qualitative valuation of results

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
28	positive	negative	positive	negative		4/4 (100%)	SFA	about the method see comments

	Sample 1	Sample 2	Sample 3	Sample 4
Spiking	positive	negative	positive	negative

Methods:

SFA = Sure Food ALLERGEN, R-Biopharm / Congen

Comments:

The results of the participant are in qualitative agreement with the spiking of samples.

Note: The method SFA indicated by the participant 28 is not known to DLA for the PCR detection of egg.

4.3 Proficiency Test Fish

4.3.1 ELISA-Results: Fish (Salmon)

Qualitative valuation of results

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
1	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	AQ	
2	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	AQ	
26	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	AQ	
17	negative	negative	negative	negative	2/4 (50%)	2/4 (50%)	BC	no positive sample deteced
34	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	BF	
5	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	EF	
28	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	EF	
23	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	IL	

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	0	7	0	7
Number negative	8	1	8	1
Percent positive	0	88	0	88
Percent negative	100	13	100	13
Consensus value	negative	positive	negative	positive
Spiking	negative	positive	negative	positive

Methods:

AQ = AgraQuant, RomerLabs

BC = BioCheck ELISA

BF = MonoTrace ELISA, BioFront Technologies

EF = SensiSpec ELISA Kit, Eurofins

IL = Immunolab

Comments:

The consensus values of results are in qualitative agreement with the spiking of samples.

One participant obtained no positive result by method BC. According to the product information of the test kit (Bio-Check) cod (100%) and other "white fish" are detected strongest. A reactivity of 8,0% is indicated for salmon which is contained in the PT-samples (s. test kit instructions).

4.3.2 PCR-Results: Fish (Salmon)

Qualitative valuation of results

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
7	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	ASU	
16	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	ASU	
32	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	ASU	
31	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	IM	
9	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	SFA	
14	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	SFA	
15	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	SFA	
24	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	SFA	
25	negative	positive	positive	negative	2/4 (50%)	2/4 (50%)	SFA	
28	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	SFA	
29	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	SFA	
33	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	SFA	
20	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	div	
21	negative	positive	negative	positive	4/4 (100%)	4/4 (100%)	div	

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	0	14	1	13
Number negative	14	0	13	1
Percent positive	0	100	7	93
Percent negative	100	0	93	7
Consensus value	negative	positive	negative	positive
Spiking	negative	positive	negative	positive

Methods:

ASU = ASU §64 Methode/method

IM = Imegen

SFA = Sure Food ALLERGEN, R-Biopharm / Congen

div = keine genaue Angabe / andere Methode

div = not indicated / other method

Comments:

The consensus values of results are in qualitative agreement with the spiking of samples.

4.4 Proficiency Test Milk

4.4.1 ELISA-Results: Milk, Casein, β-Lactoglobulin

Qualitative valuation of results

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
2	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	AQ	
10	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	AQ-P	Casein
19	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	AQ-P	Casein
34	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	BF	
5	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	EF	
28	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	EF	
18a	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	ES	Casein
18b	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	ES	β-Lactoglobulin
23	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	IL	
1	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	MI-II	Milk
21a	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	MI-II	Casein
21b	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	MI-II	β-Lactoglobulin
26a	positive	negative	positive	negative	2/4 (50%)	2/4 (50%)	MI-II	Casein
26b	positive	negative	positive	negative	2/4 (50%)	2/4 (50%)	MI-II	β-Lactoglobulin
7	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	NL-E	
16	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	RS	
25	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	RS	
4	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	RS-F	
9	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	RS-F	
12	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	RS-F	β-Lactoglobulin
14	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	RS-F	
20	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	RS-F	
27	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	RS-F	
32a	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	RS-F	
32b	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	RS-F	β-Lactoglobulin
32c	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	RS-F	Casein
6	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	VT	

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	27	0	2	25
Number negative	0	27	25	2
Percent positive	100	0	7	93
Percent negative	0	100	93	7
Consensus value	positive	negative	negative	positive
Spiking	positive	negative	negative	positive

Methods:

AQ = AgraQuant, RomerLabs
AQ-P = AgraQuant Plus, RomerLabs
BF = MonoTrace ELISA, BioFront Technologies
EF = SensiSpec ELISA Kit, Eurofins
ES = ELISA-Systems
IL = Immunolab
MI-II = Morinaga Institute ELISA Kit II
NL-E = nutriLinia®E Allergen-ELISA
RS = Ridascreen®, R-Biopharm
RS-F= Ridascreen® Fast, R-Biopharm
VT = Veratox, Neogen

Comments:
The consensus values of results are in qualitative agreement with the spiking of samples.

4.4.2 PCR-Results: Milk (Skimmed milk powder)

Qualitative valuation of results

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
28	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	SFA	about the method see comments
7	positive	negative	negative	positive	4/4 (100%)	4/4 (100%)	div	

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	2	0	0	2
Number negative	0	2	2	0
Percent positive	100	0	0	100
Percent negative	0	100	100	0
Consensus value	positive	negative	negative	positive
Spiking	positive	negative	negative	positive

Methods:

SFA = Sure Food ALLERGEN, R-Biopharm / Congen

div = keine genaue Angabe / andere Methode

div = not indicated / other method

Comments:

The results of the participants are in qualitative agreement with the spiking of samples.

The method SFA indicated by the participant 28 is not known to DLA for the PCR detection of milk.

4.5 Proficiency Test Molluscs

4.5.1 ELISA-Results: Molluscs (Squid)

Qualitative valuation of results

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
26	positive	positive	positive	positive	1/2 (50%)	2/4 (50%)	3M	
3	negative	negative	negative	negative	1/2 (50%)	2/4 (50%)	DE	no positive sample detected
5	negative	positive	negative	positive	2/2 (100%)	2/4 (50%)	EF	
21	negative	negative	negative	positive	2/2 (100%)	3/4 (75%)	EF	
28	positive	negative	negative	positive	2/2 (100%)	4/4 (100%)	EF	
23	negative	negative	negative	positive	2/2 (100%)	3/4 (75%)	IL	

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	2	2	1	5
Number negative	4	4	5	1
Percent positive	33	33	17	83
Percent negative	67	67	83	17
Consensus value	none	none	negative	positive
Spiking	positive	negative	negative	positive

Methods:

3M = 3M Protein ELISA Kit

DE = Demeditec ELISA

EF = SensiSpec ELISA Kit, Eurofins

IL = Immunolab

Comments:

The consensus values of sample 3 and 4 are in qualitative agreement with the spiking of samples. For sample 1 (lower allergen content) and sample 2 no consensus values with ≥75% positive or negative results were obtained.

Participant 26 has pointed to a possible cross-reactivity to Crustaceae for the used ELISA method 3M (see documentation). Samples 2 and 3 contain crayfish.

4.5.2 PCR-Results: Molluscs (Squid)

Qualitative valuation of results

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
12	positive	negative	negative	positive	3/3 (100%)	4/4 (100%)	4L	
7	positive	negative	negative	positive	3/3 (100%)	4/4 (100%)	SFA	
14	positive	negative	negative	positive	3/3 (100%)	4/4 (100%)	SFA	
17	negative	negative	negative	positive	3/3 (100%)	3/4 (75%)	SFA	
24	positive	negative	negative	positive	3/3 (100%)	4/4 (100%)	SFA	
25	negative	negative	negative	negative	2/3 (67%)	2/4 (50%)	SFA	no positive sample deteced
28	-	negative	negative	-	2/2 (100%)	2/2 (100%)	SFA	no positive sample deteced
31	positive	negative	negative	positive	3/3 (100%)	4/4 (100%)	SFA	
33	positive	negative	negative	positive	3/3 (100%)	4/4 (100%)	SFA	
20	negative	negative	negative	negative	2/3 (67%)	2/4 (50%)	div	no positive sample deteced

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	6	0	0	7
Number negative	3	10	10	2
Percent positive	67	0	0	78
Percent negative	33	100	100	22
Consensus value	none	negative	negative	positive
Spiking	positive	negative	negative	positive

Methods:

4L = 4LAB Diagnostics

SFA = Sure Food ALLERGEN, R-Biopharm / Congen

div = keine genaue Angabe / andere Methode

div = not indicated / other method

Comments:

The consensus values of samples 2, 3 and 4 are in qualitative agreement with the spiking of the samples. For the spiked sample 1 (lower allergen content) there were three negative results. Thus no consensus value of ≥75% positive results was obtained.

4.6 Proficiency Test Mustard

4.6.1 ELISA-Results: Mustard, in general

Qualitative valuation of results

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
2	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	AQ	
22	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	AS	Lateral Flow
10	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	BC	
34	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	BF	
28	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	EF	
23	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	IL	
32	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	NL-E	
6	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
17	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
21	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	VT	
26	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	VT	

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	11	11	11	0
Number negative	0	0	0	11
Percent positive	100	100	100	0
Percent negative	0	0	0	100
Consensus value	positive	positive	positive	negative
Spiking	positive	positive	positive	negative

Methods:

AQ = AgraQuant, RomerLabs

AS = AgraStrip (Lateral Flow), RomerLabs

BC = BioCheck ELISA

BF = MonoTrace ELISA, BioFront Technologies

EF = SensiSpec ELISA Kit, Eurofins

IL = Immunolab

NL-E = nutriLinia®E Allergen-ELISA

RS-F= Ridascreen® Fast, R-Biopharm

VT = Veratox, Neogen

Comments:

The consensus values of results are in qualitative agreement with the spiking of samples (sample 1 black, sample 2 yellow and sample 3 brown mustard).

4.6.2 PCR-Results: Mustard

Qualitative valuation of results

4.6.2.1 Mustard, in general

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
21	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	ASU	
9	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	SFA	
24	positive	positive	positive	-	3/3 (100%)	3/3 (100%)	SFA	
28	positive	positive	positive	negative	4/4 (100%)	4/4 (100%)	SFA	

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	4	4	4	0
Number negative	0	0	0	3
Percent positive	100	100	100	0
Percent negative	0	0	0	100
Consensus value	positive	positive	positive	negative
Spiking	positive	positive	positive	negative

Methods:

ASU = ASU §64 Methode/method

SFA = Sure Food ALLERGEN, R-Biopharm / Congen

Comments:

Four participants used PCR methods for the detection of mustard without differentiating the varieties. The consensus values of results are in qualitative agreement with the spiking of samples (sample 1 black, sample 2 yellow and sample 3 brown mustard).

4.6.2.2 Mustard, yellow (Sinapis alba)

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
11	negative	positive	negative	negative	4/4 (100%)	4/4 (100%)	ASU	
32	positive	positive	positive	negative	2/4 (50%)	2/4 (50%)	ASU	
14	negative	positive	negative	negative	4/4 (100%)	4/4 (100%)	CEN	
15	negative	positive	negative	negative	4/4 (100%)	4/4 (100%)	CEN	
16	negative	positive	negative	negative	4/4 (100%)	4/4 (100%)	MS	
4	negative	positive	negative	negative	4/4 (100%)	4/4 (100%)	div	
7	negative	positive	negative	negative	4/4 (100%)	4/4 (100%)	div	
20	positive	positive	positive	negative	2/4 (50%)	2/4 (50%)	div	

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	2	8	2	0
Number negative	6	0	6	8
Percent positive	25	100	25	0
Percent negative	75	0	75	100
Consensus value	negative	positive	negative	negative
Spiking	negative	positive	negative	negative

Methods:

ASU = ASU §64 Methode/method

CEN = CEN Methoden/ methods

MS = Microsynth

div = keine genaue Angabe / andere Methode

div = not indicated / other method

Comments:

Eight participants tested for mustard species by PCR. Yellow mustard (Sinapis alba) was detected in sample 2 by all of them. Two participants also obtained positive results for sample 1 and 3.

The consensus values of results are in qualitative agreement with the spiking of samples.

4.6.2.3 Mustard, brown and black (Brassica juncea / nigra)

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
32	positive	positive	positive	negative	3/4 (75%)	3/4 (75%)	ASU	
16	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	MS	
4	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	div	
7	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	div	
15	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	div	
20	positive	negative	positive	negative	4/4 (100%)	4/4 (100%)	div	

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	6	1	6	0
Number negative	0	5	0	6
Percent positive	100	17	100	0
Percent negative	0	83	0	100
Consensus value	positive	negative	positive	negative
Spiking	positive	negative	positive	negative

Methods:

ASU = ASU §64 Methode/method

MS = Microsynth

div = keine genaue Angabe / andere Methode

div = not indicated / other method

Comments:

Moreover six participants detected Brassica species in sample 1 (containing black mustard, Brassica nigra) and sample 3 (containing brown mustard, Brassica juncea). One participant also obtained positive results for sample 2.

The consensus values of results are in qualitative agreement with the spiking of samples.

4.7 Proficiency Test Soya

4.7.1 ELISA-Results: Soya (Soyflour)

Qualitative valuation of results

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
2	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	AQ	
22	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	AS	Lateral Flow
10	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	BC	
34	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	BF	
28	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	EF	
18	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	ES	
23	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	IL	
1	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	MI-II	
21	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	MI-II	
26	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	MI-II	
4	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
5	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
6	negative	positive	positive	positive	3/4 (75%)	3/4 (75%)	RS-F	
7	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
8	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	Sample 1 and 4: Traces below LOD
14	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
19	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
20	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
32	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	RS-F	
17	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	VT	

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	0	20	20	1
Number negative	20	0	0	19
Percent positive	0	100	100	5
Percent negative	100	0	0	95
Consensus value	negative	positive	positive	negative
Spiking	negative	positive	positive	negative

Methods:

AQ = AgraQuant, RomerLabs

AS = AgraStrip (Lateral Flow), RomerLabs

BC = BioCheck ELISA

BF = MonoTrace ELISA, BioFront Technologies

EF = SensiSpec ELISA Kit, Eurofins

ES = ELISA-Systems

IL = Immunolab

MI-II = Morinaga Institute ELISA Kit II

RS-F= Ridascreen® Fast, R-Biopharm

VT = Veratox, Neogen

Comments:

The consensus values of results are in qualitative agreement with the spiking of samples.

4.7.2 PCR-Results: Soya (Soyflour)

Qualitative valuation of results

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Qualitative Valuation	Qualitative Valuation	Method	Remarks
	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples		
11	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	ASU	
32	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	ASU	
4	negative	positive	negative	negative	3/4 (75%)	3/4 (75%)	MS	
16	positive	positive	positive	negative	3/4 (75%)	3/4 (75%)	MS	
9	negative	negative	positive	positive	2/4 (50%)	2/4 (50%)	SFA	
25	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	SFA	
28	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	SFA	
7	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	div	
8	negative	positive	negative	negative	3/4 (75%)	3/4 (75%)	div	Sample 3 traces below LOD
15	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	div	
20	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	div	
21	negative	positive	positive	negative	4/4 (100%)	4/4 (100%)	div	

	Sample 1	Sample 2	Sample 3	Sample 4
Number positive	1	11	10	1
Number negative	11	1	2	11
Percent positive	8	92	83	8
Percent negative	92	8	17	92
Consensus value	negative	positive	positive	negative
Spiking	negative	positive	positive	negative

Methods:

ASU = ASU §64 Methode/method

MS = Microsynth

SFA = Sure Food ALLERGEN, R-Biopharm / Congen

div = keine genaue Angabe / andere Methode

div = not indicated / other method

Comments:

The consensus values of results are in qualitative agreement with the spiking of samples.

5. Documentation

5.1 Details by the participants

Note: Information given in German was translated by DLA to the best of our knowledge (without guarantee of correctness).

5.1.1 ELISA: Crustaceae

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
AQ	1		negative	positive	positive	negative	0,1	Please select!	AQ = AgraQuant, RomerLabs
AQ	2	19/08	negative	positive	positive	negative	0,02	Crustacea Protein	AQ = AgraQuant, RomerLabs
AQ	17	01.07.19	pos	pos	neg	neg	0,1	Crustacea	Romer
AQ	26	07.12.19	negative	positive	positive	negative	0,02	tropomyosine	AQ = AgraQuant, RomerLabs
BF	24		negative	positive	positive	negative	1	Food item, total	BF = MonoTrace ELISA, BioFront Technologies
BF	34	09.10.19	negative	positive	positive	negative	0,07	Food item, total	BF = MonoTrace ELISA, BioFront Technologies
EF	5	30.08.19	negative	positive	positive	negative	0,0009	Food item, total	EF = SensiSpec ELISA Kit, Eurofins
EF	21	15.07.19	negative	positive	positive	negative	0,01	Please select!	EF = SensiSpec ELISA Kit, Eurofins
EF	28		negative	positive	positive	negative	0,02	Please select!	EF = SensiSpec ELISA Kit, Eurofins
EF	30	07.10.19	negative	positive	positive	negative	0,02	Food item, fresh mass	EF = SensiSpec ELISA Kit, Eurofins
ES	14	16/July	negative	0,36	0,18	negative	0,05	protein	ES = ELISA-Systems
IL	23	24.07.19	negative	positive	positive	negative			IL = Immunolab
NL-E	7	29.08.19	negative	positive	positive	negative	0,001	Tropomyosin	NL-E = nutriLinia®E Allergen-ELISA
RS-F	3	23.07.19	negative	positive	positive	negative	20	Food item, total	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	6		positive	positive	positive	positive	2	Please select!	r-Biopharm R7312
RS-F	12	26.07.19	negative	positive	positive	negative	2	Food item, total	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	20	23.07.19	neg	pos	pos	neg	2	Protein	R-BIOPHARM R7312
RS-F	32	16.07.19	negative	positive	positive	negative	2	Food item, total	RS = Ridascreen®, R-Biopharm
VT	13	23.07.19	negative	positive	positive	negative	2,5	Food item, total	VT = Veratox, Neogen

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Antibody	e.g. Extractionbuffer / Time / Temperature	
AQ	1	COKAL2248 Kit Lot# CR101701804			Detection gven as: Crustacea protein
AQ	2				
AQ	17	COKAL2248		Detects Tropomysin, converted to 'Crustacea'.	
AQ	26	COKAL2248		as stipulated in kit insert	kit unit: ppb tropomyosine, converted to ppm
BF	24				
BF	34		Monoclonal; anti-tropomyosin	1:10 extraction ration	
EF	5	HU0030006,HU0030030			
EF	21	HU0030006	detects crustacean tropomyosin	according to manufacturer's instructions	NWG indicated as tropomyosin crustaceans
EF	28				
EF	30	HU0030006			
ES	14	ESCRURD-48 (CRU18-337)	Tropomyosin	Allergen extraction buffer (Kit)/30-15-10/ room temperature	
IL	23	CRU-E01	Tropomyosin		
NL-E	7	NC-6051		according to instructions	
RS-F	3	R7312	As Per Kit Instructions	As Per Kit Instructions	
RS-F	6	r-Biopharm R6152			
RS-F	12	R7312	CRUSTACEAN PROTEIN	ONE BUFFER EXTRACTION (60°C, 10 MIN)	
RS-F	20				
RS-F	32	R7312	Tropomyosin	Extraktionspuffer/10 min/ 60°C	
VT	13				

5.1.2 ELISA: Egg

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
AQ	2	29/07	positive	negative	positive	negative	0,4	Egg White Protein	AQ = AgraQuant, RomerLabs
AQ	4	30.08.19	positive	negative	positive	negative	0,05	Egg white protein	AQ
BC	10	18.07.19	positive	negative	positive	negative	0,4	Food item, total	BC = BioCheck ELISA
BF	34	09.10.19	positive	negative	positive	negative	0,3	Whole egg powder	BF = MonoTrace ELISA, BioFront Technologies
BK	6		positive	negative	positive	negative	0,1	Please select!	Neogen Biokits 902072T
EF	5	30.08.19	positive	negative	positive	negative	0,05	Egg white powder	EF = SensiSpec ELISA Kit, Eurofins
EF	28		positive	negative	positive	negative	10	Please select!	EF = SensiSpec ELISA Kit, Eurofins
ES	18		positive	negative	positive	negative	5 ppm	Egg white powder	ES = ELISA-Systems
IL	23	23.07.19	positive	negative	positive	negative			IL = Immunolab
MI	1		positive	negative	positive	negative	0,3	Please select!	MI = Morinaga Institute ELISA
MI	26	31.07.19	positive	negative	positive	negative	0,31	Protein, total	MI = Morinaga Institute ELISA
MI-II	16		positive	positive	positive	positive	10	Please select!	Auswahl ELISA-Kits:
MI-II	21	15.07.19	positive	negative	positive	negative	0,31	Please select!	MI-II = Morinaga Institute ELISA Kit II
MI-II	24		positive	negative	positive	negative		Whole egg powder	MI-II = Morinaga Institute ELISA Kit II
RS	25		positive	negative	positive	negative	0,1	Please select!	RS = Ridascreen®, R-Biopharm
RS-F	7b	29.08.19	positive	negative	positive	negative	0,1	Whole egg powder	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	9	22.08.19	positive	negative	positive	negative	0,03 mg/kg Eiklar-Protein	Food item, total	Ridascreen Fast Ei /SFA-Q
RS-F	12	25.07.19	positive	negative	positive	negative	0,1	Whole egg powder	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	14	17/July	>13,5	negative	>13,5	negative	0,5	food/food	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	17	01.07.19	pos	neg	pos	neg	0,5	Whole Egg Powder	R-Biopharm
RS-F	19	15.07.19	positive	negative	positive	negative	0,3	Egg white powder	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	20	16.07.19	pos	neg	pos	neg	0,05	Protein	R-BIOPHARM 6402
RS-F	32	15.07.19	positive	negative	positive	negative	0,1	Whole egg powder	RS-F= Ridascreen® Fast, R-Biopharm
RS-L	7a	29.08.19	positive	negative	positive	negative	0,02	Lysozyme	RS = Ridascreen®, R-Biopharm

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Antibody	e.g. Extractionbuffer / Time / Temperature	
AQ	2				
AQ	4	COKAL0848	Egg white protein	according to kit instructions	Sample 1 =18,5 mg/kg; Sample 3 = 27,0 mg/kg
BC	10	Kit No 12811			
BF	34		Monoclonal; anti-ovomucoid	1:20 extraction ratio	
BK	6				
EF	5	HU0030007,HU0030031			
EF	28				
ES	18	ES-6020, Transia	polyclonal, anti Ovomucoid/Ovalbumin	Extraction buffer, 3x10min, room temperature	LOD 0,5 ppm
IL	23	EGG-E01	Ovomucoid		
MI	1	M2101 - Kit Lot#1901SAOA147			Detection gven as: Egg protein
MI	26	M2101		overnight extraction, room temperature	kit unit: ppm egg protein
MI-II	16	MI-II		according to kit instructions	
MI-II	21	M2111	Detects egg white protein Ovalbumin	according to manufacturer's instructions	LOD given as whole egg protein
MI-II	24				
RS	25				
RS-F	7b	R-6402		according to kit instructions	
RS-F	9	R6402	specific egg white proteins ovalbumin and ovomucoid	Extraction solution 60°C	
RS-F	12	R6402	EGG PROTEIN	ONE BUFFER EXTRACTION (60°C, 10 MIN)	
RS-F	14	R6402 (15358)	unknown	Allergen extraction buffer (Rbiopharm)/10-10-10/ room temperature	
RS-F	17	R6402		Detects egg white proteins, calibrated against whole egg powder	
RS-F	19	R6402			
RS-F	20				
RS-F	32	R6402	Ovalbumin	Extraction buffer/10 min/ 60°C	
RS-L	7a	R6452		according to instructions	

5.1.3 ELISA: Fish

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
AQ	1		negative	positive	negative	positive	4	Please select!	AQ = AgraQuant, RomerLabs
AQ	2	08.07.19	negative	positive	negative	positive	4	Protein, Total	AQ = AgraQuant, RomerLabs
AQ	26	07.12.19	negative	positive	negative	positive	4	cod	AQ = AgraQuant, RomerLabs
BC	17	01.07.19	neg	neg	neg	neg	5	Fish (Cod Parvalbumin)	Biocheck-UK
BF	34	09.10.19	negative	positive	negative	positive	0,8	Food item, total	BF = MonoTrace ELISA, BioFront Technologies
EF	5	30.08.19	negative	positive	negative	positive	1,4	Food item, total	EF = SensiSpec ELISA Kit, Eurofins
EF	28		negative	positive	negative	positive	4	Please select!	EF = SensiSpec ELISA Kit, Eurofins
IL	23	24.07.19	negative	positive	negative	positive			IL = Immunolab

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Antibody	e.g. Extractionbuffer / Time / Temperature	
AQ	1	COKAL2548 Kit lot# FI1022-1807			Detection given as Cod protein
AQ	2				
AQ	26	COKAL2548		as stipulated in kit insert	kit unit: ppm cod
BC	17	R6009		Detects cod-parvalbumin 100%, lesser reactivity to other fish	
BF	34		Monoclonal; anti-tropomyosin	1:10 extraction ration	
EF	5	HU0030008,HU0030032			
EF	28				
IL	23	FIS-E01	Parvalbumin		

5.1.4 ELISA: Milk

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
AQ	2	23/07	positive	negative	negative	positive	0,4	Protein, Total	AQ = AgraQuant, RomerLabs
AQ-P	10	23.07.19	positive	negative	negative	positive	0,2	Food item, total	AQ-P = AgraQuant Plus, RomerLabs
AQ-P	19	18.07.19	positive	negative	negative	positive	1,5	Food item, total	AQ-P = AgraQuant Plus, RomerLabs
BF	34	09.10.19	positive	negative	negative	positive	0,12	Egg white powder	BF = MonoTrace ELISA, BioFront Technologies
EF	5	30.08.19	positive	negative	negative	positive	0,05	Food item, total	EF = SensiSpec ELISA Kit, Eurofins
EF	28		positive	negative	negative	positive	0,4	Please select!	EF = SensiSpec ELISA Kit, Eurofins
ES	18a		positive	negative	negative	positive	10 ppm Casein; 1 ppm ß-Lactglobulin	Skimmed milk powder equivalents or ß-Lactglobulin	ES = ELISA-Systems
ES	18b		positive	negative	negative	positive	10 ppm Casein; 1 ppm ß-Lactglobulin	Skimmed milk powder equivalents or ß-Lactglobulin	ES = ELISA-Systems
IL	23	24.07.19	positive	negative	negative	positive			IL = Immunolab
MI-II	1		positive	negative	negative	positive	0,3	Please select!	MI-II = Morinaga Institute ELISA Kit II
MI-II	21a	12.07.19	positive	negative	negative	positive	0,31	Please select!	MI-II = Morinaga Institute ELISA Kit II
MI-II	21b	12.07.19	positive	negative	negative	positive	0,31	Please select!	MI-II = Morinaga Institute ELISA Kit II
MI-II	26a	16.07.19	positive	negative	positive	negative	0,31	Protein, total	MI-II = Morinaga Institute ELISA Kit II
MI-II	26b	16.07.19	positive	negative	positive	negative	0,31	Protein, total	MI-II = Morinaga Institute ELISA Kit II
NL-E	7	29.08.19	positive	negative	negative	positive	0,1	Milk protein	NL-E = nutriLinia®E Allergen-ELISA
RS	16		positive	negative	negative	positive	10	Please select!	Auswahl ELISA-Kits:
RS	25		positive	negative	negative	positive	1,5	Please select!	RS = Ridascreen®, R-Biopharm
RS-F	4	23.08.19	positive	negative	negative	positive	0,7 mg/kg	Milk protein	RS-F
RS-F	9	21.08.19	positive	negative	negative	positive	0,7 mg/kg Milchprotein	Food item, total	Ridascreen Fast Milk/SFA-Q
RS-F	12	30.07.19	positive	negative	negative	positive	0,04	Food item, total	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	14	15/July	24,3	negative	negative	19,6	2,5	protein	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	20	01.08.19	pos	neg	neg	pos	0,7	Protein	R-BIOPHARM 4652
RS-F	27	30.07.19	positive	negative	negative	positive	0,7	Milk powder	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	32a	12.07.19	positive	negative	negative	positive	0,7	Milk powder	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	32b	12.07.19	positive	negative	negative	positive	0,04	Food item, total	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	32c	12.07.19	positive	negative	negative	positive	0,7	Food item, total	RS-F= Ridascreen® Fast, R-Biopharm
VT	6		positive	negative	negative	positive	1	Please select!	Neogen Veratox 8470

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Antibody	e.g. Extractionbuffer / Time / Temperature	
AQ	2				
AQ-P	10	Kit No CA1041-1903			
AQ-P	19	COKAL248F			tested for casein
BF	34		Monoclonal; anti-casein	1:10 extraction ratio	
EF	5	HU0030014, HU0030038			
EF	28				
ES	18a	ES-6030 Transia, ES-6034 Transia	polyclonal, anti bovine alpha-Casein or anti ß-Lactglobulin	for both ELISAs extraction buffers, 2x15min and 1x10 min, room temperature	w e use 2 ELISAs: LOD 1 ppm skimmed milk pow der equivalents or LOD 0,1 ppm ß-Lactglobulin
ES	18b	ES-6030 Transia, ES-6034 Transia	polyclonal, anti bovine alpha-Casein or anti ß-Lactglobulin	for both ELISAs extraction buffers, 2x15min and 1x10 min, room temperature	w e use 2 ELISAs: LOD 1 ppm skimmed milk pow der equivalents or LOD 0,1 ppm ß-Lactglobulin
IL	23	MIL-E01	Casein, ß-Lactoglobulin		
MI-II	1	M2112 19JASFBL029 M2114 19MASFCS070			Detection gven as: w hole milk protein. Positive for BLG and Casein
MI-II	21a	M2113	Detects cow milk Casein	according to manufacturer's instructions	LOD given as milk protein
MI-II	21b	M2112	Detects cow milk ß-Lactoglobulin	according to manufacturer's instructions	LOD given as milk protein
MI-II	26a	M2113		overnight extraction, room temperature	Casein ELISA kit II kit unit: ppm milkprotein
MI-II	26b	M2112		overnight extraction, room temperature	BLG ELISA kit II kit unit: ppm milkprotein
NL-E	7	NC-6033		according to the manual	
RS	16	RS		accoding to the kit manual	
RS	25				
RS-F	4	R4652	Casein and ß-Lactoglobulin	according to kit manual	sample 1 =36,3 mg/kg; sample 4 = 12,4 mg/kg
RS-F	9	R4652	specific f. caseine of cow , sheep, goat, buffalo	Extraction solution 60 °C, Extraktor 2 100 °C	
RS-F	12	R4912	COW BETALACTOGLOBULIN	TWO BUFFERS EXTRACTION (60°C, 10 MIN-100°C, 10 MIN)	
RS-F	14	R4652 (15249)	unknow n	Allergen extraction buffer (Rbiopharm)/10-10-10/ room temperature	
RS-F	20				
RS-F	27	R4652	see kit manual	Processing of the sample exactly as specified by the manufacturer	
RS-F	32a	R4652	Casein / b-Lactoglobulin	AAEP/100°C_10 min/AEP/10 min/ 60°C_ dilution 1:5	
RS-F	32b	R4912	b-Lactoglobulin	AAEP/100°C_10 min/AEP/10 min/ 60°C_ dilution 1:5	
RS-F	32c	R4612	Casein	AAEP/100°C_10 min/AEP/10 min/ 60°C_ dilution 1:5	
VT	6				

5.1.5 ELISA: Molluscs

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
3M	26	26.07.19	positive	positive	positive	positive	1	Protein, total	3M = 3M Protein ELISA Kit
DE	3	23.07.19	negative	negative	negative	negative	0,01	Please select!	DE = Demeditec ELISA
EF	5	30.08.19	negative	positive	negative	positive	1,7	Food item, total	EF = SensiSpec ELISA Kit, Eurofins
EF	21	15.07.19	negative	negative	negative	positive	0,01	Please select!	EF = SensiSpec ELISA Kit, Eurofins
EF	28		positive	negative	negative	positive	0,01	Please select!	EF = SensiSpec ELISA Kit, Eurofins
IL	23	24.07.19	negative	negative	negative	positive			IL = Immunolab

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Antibody	e.g. Extractionbuffer / Time / Temperature	
3M	26	E96MOL		as stipulated in kit insert	kit unit: ppm molluscs protein Sample 2 and 3 w ere found positive to crustaceae. Cross reactivity to crustaceae is know n w ith 3M Mollusk Protein ELISA Kit
DE	3	DEMOLE01	As Per Kit Instructions	As Per Kit Instructions	LOD as tropomyosin
EF	5	HU0030015,HU0030039			
EF	21	HU0030015	Detects mollusc tropomyosin	according to manufacturer's instructions	LOD given as tryopomyosin molluscs
EF	28				
IL	23	MOL-E01	Tropomyosin		

5.1.6 ELISA: Mustard

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detec-tion given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
AQ	2	08.07.19	positive	positive	positive	negative	2	Food item, total	AQ = AgraQuant, RomerLabs
AS	22	15.06.19	positive	positive	positive	negative	2		AgraStrip Mustard / Romer Labs
BC	10	24.07.19	positive	positive	positive	negative	2	Food item, total	BC = BioCheck ELISA
BF	34	09.10.19	positive	positive	positive	negative	0,13	Food item, total	BF = MonoTrace ELISA, BioFront Technologies
EF	28		positive	positive	positive	negative	2	Please select!	EF = SensiSpec ELISA Kit, Eurofins
IL	23	23.07.19	positive	positive	positive	negative			IL = Immunolab
NL-E	32	17.07.19	positive	positive	positive	negative	1	Food item, total	NL-E = nutriLinia®E Allergen-ELISA
RS-F	6		positive	positive	positive	negative	0,11	Please select!	r-Biopharm R6152
RS-F	17	01.07.19	pos	pos	pos	neg	0,5	Mustard	R-Biopharm
VT	21	12.07.19	positive	positive	positive	negative	1,5	Please select!	BK = BioKits, Neogen
VT	26	26.07.19	positive	positive	positive	negative	2,5	Food item, total	VT = Veratox, Neogen

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Antibody	e.g. Extractionbuffer / Time / Temperature	
AQ	2				
AS	22	COKAL2110AS			
BC	10	Kit No 12726			
BF	34		Monoclonal; anti-Sin a 1	1:20 extraction ratio; assay cannot differentiate betw een yellow , brow n, or black mustard	
EF	28				
IL	23	MUS-E01	Mustard, total		
NL-E	32	NC-6007	Mustard seed proteins	Extraction buffer/15 min/ 60°C	
RS-F	6			In the indication of results, no distinction is made in mustard yellow / w hite, brow n or black. It is about "total mustard"	
RS-F	17	R6152		Detects all mustard types and reports as 'mustard'.	
VT	21	8400	detects mustard protein from seeds of w hite, black and brow n mustard	according to manufacturer's instructions	LOD given as mustard
VT	26	8400		as stipulated in kit insert	kit unit: ppm mustard seeds Veratox for Mustard Allergen do not distinguish mustard varieties.

5.1.7 ELISA: Soya

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
AQ	2	23/07/19	negative	positive	positive	negative	0,3	Protein, Total	AQ = AgraQuant, RomerLabs
AS	22	15.06.19	negative	positive	positive	negative	2		AgraStrip Soy / Romer Labs
BC	10	19.07.19	negative	positive	positive	negative	10	Please select!	BC = BioCheck ELISA
BF	34	09.10.19	negative	positive	positive	negative	0,16	Food item, total	BF = MonoTrace ELISA, BioFront Technologies
EF	28		negative	positive	positive	negative	0,04	Please select!	EF = SensiSpec ELISA Kit, Eurofins
ES	18		negative	positive	positive	negative	25 ppm	Soyprotein	ES = ELISA-Systems
IL	23	24.07.19	negative	positive	positive	negative			IL = Immunolab
MI-II	1		negative	positive	positive	negative	0,3	Please select!	MI-II = Morinaga Institute ELISA Kit II
MI-II	21	15.07.19	negative	positive	positive	negative	0,31	Please select!	MI-II = Morinaga Institute ELISA Kit II
MI-II	26	17.07.19	negative	positive	positive	negative	0,31	Protein, total	MI-II = Morinaga Institute ELISA Kit II
RS-F	4	27.08.19	negative	positive	positive	negative	0,24 mg/kg	Soyprotein	RS-F
RS-F	5	30.08.19	negative	positive	positive	negative	0,31	Please select!	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	6		negative	positive	positive	positive	0,24	Please select!	r-Biopharm R7102
RS-F	7	29.08.19	negative	positive	positive	negative	0,31	Soyprotein	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	8	22-23/8/19	negative (<LOQ)	positive	positive	negative (<LOQ)	0,24 ppm soy protein (LOD) 2,5 ppm soy protein (LOQ)	Food item, dry mass	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	14	15/July	negative	>20	>20	negative	2,5	protein	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	19	16.07.19	negative	positive	positive	negative	4	Food item, total	RS-F= Ridascreen® Fast, R-Biopharm
RS-F	20	23.07.19	neg	pos	pos	neg	0,24	Protein	R-BIOPHARM 7102
RS-F	32	16.07.19	negative	positive	positive	negative	0,24	Food item, total	RS-F= Ridascreen® Fast, R-Biopharm
VT	17	01.07.19	neg	pos	pos	neg	10	Soy	Veratox

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Antibody	e.g. Extractionbuffer / Time / Temperature	
AQ	2				
AS	22	COKAL0410AS			
BC	10	Kit No 12675			Expressed as Roasted Soya Protein
BF	34		Monoclonal; anti-Gly m 6	1:20 extraction ratio, boiling for 10 minutes	
EF	28				
ES	18	ES-6012, Transia	polyclonal, anti-soya trypsin Inhibitor and anti-soyflour protein	Extraction buffer, 2x30min and 1x15 min, room temperature	LOD 2,5 ppm
IL	23	SOJ-E01	Soy trypsin inhibitor		
MI-II	1	M2177 Kit Lot# 19MASFSY032			Detection gven as: Whole Soya protein
MI-II	21	M2117	detects the soy protein Beta-Conglycinin	according to manufacturer's instructions	LOD given as soy protein
MI-II	26	M2117		short extraction, 100°C	kit unit: ppm soy protein
RS-F	4	R7102	soyprotein	according to kit manual	sample 2 = 8,1 mg/kg, sample 3 = 4,1 mg/kg
RS-F	5	R 7102			in soy protein
RS-F	6				
RS-F	7	R7102		according to manual	
RS-F	8	R7102, 13569/LT	antibodies specifically detect heated soy protein	protocol 9.1 (solid samples) w as used for protein extraction	NT: Not Tested; Samples 1 and 4 are >LOD but <LOQ thus not quantifiable; Samples 2 and 3 w ere >LOD and >LOQ and w ere also quantified (sample 2: 51,83 mg/kg soy protein; sample 3: 23,04 mg/kg soy protein)
RS-F	14	R7102 (13428)	unknow n	Allergen extraction buffer (Rbiopharm)/10-10-10/ room temperature	
RS-F	19	R7102			
RS-F	20				
RS-F	32	R7102	Soy proteins, polyclonal	Extractor +Extraction buffer/10 min/100°C; dil. 1:5	
VT	17	8410		Soy Flour	

5.1.8 PCR: Crustaceae

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
SFA	3	19.08.19	negative	positive	positive	negative	1	Food item, total	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	7	29.08.19	negative	positive	positive	negative	0,4	Giant prawn tails	SFA-Q = Sure Food Allergen Quant, R-Biopharm / Congen
SFA	14	12/July	negative	positive	positive	negative	1	food/food	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	24		negative	positive	positive	negative	0,4	Allergen DNA	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	28		negative	positive	positive	negative	0,4	Please select!	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	29		negative	positive	positive	negative		Allergen DNA	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	33		neg	pos	pos	neg	50	Food	R Biopharm CONGEN Kit

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Target-DNA	e.g. Extraction / Enzymes / Clean-Up / Real Time PCR / Gel electrophoresis / Cycles	
SFA	3	S3612	As Per Kit Instructions	As Per Kit Instructions	
SFA	7	S3612		according to manual	
SFA	14	S3612 (12258)	unknow n	Extraction: NucleoSpin Food (Macherey Nagel)/Real Time PCR / 45 cycles	
SFA	24				
SFA	28				
SFA	29	S3612			
SFA	33	S3112		In house exatrction method. Phenol/Chloroform follow ed by Qiagen DNEasy Plant kit. Real time PCR - 35 cycles	

5.1.9 PCR: Egg

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
SFA	28		positive	negative	positive	negative	0,4	Please select!	SFA = Sure Food ALLERGEN, R-Biopharm / Congen

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Target-DNA	e.g. Extraction / Enzymes / Clean-Up / Real Time PCR / Gel electrophoresis / Cycles	
SFA	28				

5.1.10 PCR: Fish

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
ASU	7	29.08.19	negative	positive	negative	positive	5 copies	Food item, total	ASU = ASU §64 Methode/method
ASU	16		negative	positive	negative	positive		Please select!	Selection PCR methods
ASU	32		negative	positive	negative	positive	10	Food item, total	ASU = ASU §64 Methode/method
IM	31		negative	positive	negative	positive	0,4	Food item, total	Other: Imegen
SFA	9	20.08.19	negative	positive	negative	negative		Allergen-DNA	SFA-ID = Sure Food Allergen ID, R-Biopharm / Congen
SFA	14	12/July	negative	positive	negative	positive	1	food/food	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	15		negative	positive	negative	positive	2,5	Food item, total	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	24		negative	positive	negative	positive	0,4	Allergen DNA	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	25		negative	positive	positive	negative		Please select!	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	28		negative	positive	negative	positive	1	Please select!	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	29		negative	positive	negative	positive		Allergen DNA	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	33		neg	pos	neg	pos	100	Food	R Biopharm CONGEN Kit
div	20		neg	pos	neg	pos	8		in-house method
div	21	12.07.19	negative	positive	negative	positive	20	Allergen-DNA	Selection PCR methods

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Target-DNA	e.g. Extraction / Enzymes / Clean-Up / Real Time PCR / Gel electrophoresis / Cycles	
ASU	7		Hoxc 13 Gen	Real Time PCR, 45 cycles	
ASU	16	ASU		Wizard Promega	
ASU	32	L10.00-12 / 2012-07	cyt b / Parvalbumin	CTAB, Proteinase K/60°C/ Clean up: Dneasy Mericon Food Kit (Qiagen)	
IM	31			CTAB/ kit /PCR real time	
SFA	9	S3610		SureFood® PREP Advanced	
SFA	14	S3610 (15238)	unknow n	Extraction: NucleoSpin Food (Macherey Nagel)/Real Time PCR / 45 cycles	
SFA	15	S3610		Extraction CTAB; real time PCR, 45 cycles	
SFA	24				
SFA	25				
SFA	28				
SFA	29	S3610			
SFA	33	S3610		In house exatrction method. Phenol/Chloroform follow ed by Qiagen DNEasy Plant kit. Real time PCR - 50 cycles	
div	20				Limit of detection given as µg of DNA per kg of sample
div	21	internal method		CTAB / Proteinase K / Promega Wizard DNA-CleanUp / Real-time PCR / 45 cycles	

5.1.11 PCR: Milk

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
SFA	28		positive	negative	negative	positive	0,4	Please select!	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
div	7	29.08.19	positive	negative	negative	positive	< 1 copies	Food item, total	in house method

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Target-DNA	e.g. Extraction / Enzymes / Clean-Up / Real Time PCR / Gel electrophoresis / Cycles	
SFA	28				
div	7		mitochondrial	Real Time PCR, 45 cycles	

5.1.12 PCR: Molluscs

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
4L	12	07.08.19	positive	negative	negative	positive	1,6 pg target DNA /100 ng total DNA	Allergen DNA	4L = 4LAB Diagnostics
SFA	7	29.08.19	positive	negative	negative	positive	0,4	Food item, total	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	14	12/July	positive	negative	negative	positive	1	food/food	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	17	01.07.19	neg	neg	neg	pos	1	Molluscs	SureFood R-Biopharm (Congen)
SFA	24		positive	negative	negative	positive	0,4	Allergen DNA	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	25		negative	negative	negative	negative		Please select!	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	28		-	negative	negative	-	0,4	Please select!	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	31		positive	negative	negative	positive	100	Food item, total	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	33		pos	neg	neg	pos	50	Food	R Biopharm CONGEN Kit
div	20		neg	neg	neg	neg	80		in-house method

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Target-DNA	e.g. Extraction / Enzymes / Clean-Up / Real Time PCR / Gel electrophoresis / Cycles	
4L	12	IC-02-1008	MOLLUSC DNA	GREES DNA FOOD KIT IC-02-0095 (LYSIS SOLUTION, EXTRACTION COLUMNS)	
SFA	7	S3613		according to manual	
SFA	14	S3613 (14059)	unknown	Extraction: NucleoSpin Food (Macherey Nagel)/Real Time PCR / 45 cycles	
SFA	17	S3613		Tris extraction column clean up	
SFA	24				
SFA	25				
SFA	28				
SFA	31			CTAB/ kit /PCR real time	
SFA	33	S3113		In house exatrction method. Phenol/Chloroform followed by Qiagen DNEasy Plant kit. Real time PCR - 35 cycles	
div	20				Limit of detection given as µg of DNA per kg of sample

5.1.13 PCR: Mustard, in general

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
ASU	21	12.07.19	positive	positive	positive	negative	10	Allergen-DNA	ASU = ASU §64 Methode/method
SFA	9	23.07.19	positive	positive	positive	negative		Allergen-DNA	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	24		positive	positive	positive	-	0,4	Allergen DNA	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	28		positive	positive	positive	negative	0,4	Please select!	SFA = Sure Food ALLERGEN, R-Biopharm / Congen

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Target-DNA	e.g. Extraction / Enzymes / Clean-Up / Real Time PCR / Gel electrophoresis / Cycles	
ASU	21	L 08.00-65:2017-10		CTAB / Proteinase K / Promega Wizard DNA-CleanUp / Real-time PCR / 45 cycles	
SFA	9	S3609	DNA from brown, yellow and black mustard	SureFood® PREP Advanced	
SFA	24				The kit used for mustard's determination detects all three species listed without distinction.
SFA	28				

5.1.14 PCR: Mustard, Sinapis alba

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
ASU	11	02.08.19	negative	positive	negative	negative	10	Food item, total	ASU
ASU	32	18.07.19	positive	positive	positive	negative	4	Food item, total	ASU = ASU §64 Methode/method
CEN	14	26/July	negative	positive	negative	negative	1	food/food	UNE-CEN/TS 15634-5:2016
CEN	15		negative	positive	negative	negative	5	Food item, total	Selection PCR-Methods
MS	16		negative	positive	negative	negative		Please select!	Selection PCR-Methods
div	4	27.08.19	negative	positive	negative	negative	100	Mustard-DNA	in-house method
div	7	29.08.19	negative	positive	negative	negative	< 10 copies	Food item, total	in-house method
div	20		pos	pos	pos	neg	8		in-house method

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Target-DNA	e.g. Extraction / Enzymes / Clean-Up / Real Time PCR / Gel electrophoresis / Cycles	
ASU	11	L08.00-59	MADS D-F, MADSD-R	CTAB with / without precipitation, Dneasy Mericon Food	
ASU	32	L08.00-59/2013-01	MADS	CTAB, Proteinase K/60°C/ Clean up: Dneasy Mericon Food Kit (Qiagen)	
CEN	14		MADS-D	Extraction: NucleoSpin Food (Macherey Nagel)/Real Time PCR / 45 cycles	
CEN	15	UNE CEN/TS 15634-5	74 pb	Extraction CTAB; real time PCR multiplex, 50 cycles	Sonda and primers for detection white Sinapis alba, and sonda y primers for detection brown/black Brassica nigra/Brassica juncea
MS	16	MS		Wizard Promega	
div	4		Sinapis alba	DNA Extraction with Proteinase K, Clean Up with chloroform and columns /Amplif m RealTIme PCR 45 cycles	
div	7		MADS-D protein	Real Time PCR, 45 cycles	
div	20			our detection method targets Sinapis alba	Limit of detection given as µg of DNA per kg of sample

5.1.15 PCR: Mustard, Brassica juncea/ Brassica nigra

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
ASU	32	18.07.19	positive	positive	positive	negative	4	Food item, total	ASU = ASU §64 Methode/method
MS	16		positive	negative	positive	negative		Please select!	Selection PCR-Methods
div	4	27.08.19	positive	negative	positive	negative	100	Mustard-DNA	Hausmethode
div	7	29.08.19	positive	negative	positive	negative	< 5 copies	Food item, total	Hausmethode
div	15		positive	negative	positive	negative	5	Food item, total	Selection PCR-Methods
div	20		pos	neg	pos	neg	8		in-house method

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Target-DNA	e.g. Extraction / Enzymes / Clean-Up / Real Time PCR / Gel electrophoresis / Cycles	
ASU	32	L08.00-64/2016-10		CTAB, Proteinase K/60°C/ Clean up: Dneasy Mericon Food Kit (Qiagen)	
MS	16	MS		Wizard Promega	
div	4		Brassica juncea/nigra	DNA extraction w ith proteinase K, Clean Up w ith chloroform and columns /Amplif m RealTIme PCR 45 cycles	no distinction betw een juncea / nigra possible
div	7		Gypsy-like retro element	Real Time PCR, 45 cycles	Method can not differentiate betw een brow n and black mustard
div	15	Palle Reich et al. (2013). Food Chemistry	76 pb	Extraction CTAB; real time PCR multiplex, 50 cycles	Sonda and primers for detection w hite Sinapis alba, and sonda y primers for detection brow n/black Brassica nigra/Brassica juncea
div	20			our detection method targets Brassica juncea/nigra	Limit of detection given as µg of DNA per kg of sample

5.1.16 PCR: Soya

Primary data

Meth. Abr.	Evaluation number	Date of analysis	Result Sample 1	Result Sample 2	Result Sample 3	Result Sample 4	Limit of detection	Limit of detection given as	Method
		Day/ Month	positive / negative	positive / negative	positive / negative	positive / negative	mg/kg	e.g. food / food protein	Test-Kit + Provider
ASU	11	02.08.19	negative	positive	positive	negative	10	Food item, total	ASU
ASU	32	18.07.19	negative	positive	positive	negative	4	Food item, total	ASU = ASU §64 Methode/method
MS	4	28.08.19	negative	positive	negative	negative	50	Soya DNA	MS
MS	16		positive	positive	positive	negative		Please select!	Selection PCR-Methods
SFA	9	22.07.19	negative	negative	positive	positive		Allergen-DNA	SFA-ID = Sure Food Allergen ID, R-Biopharm / Congen
SFA	25		negative	positive	positive	negative		Please select!	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
SFA	28		negative	positive	positive	negative	0,4	Please select!	SFA = Sure Food ALLERGEN, R-Biopharm / Congen
div	7	29.08.19	negative	positive	positive	negative	< 10 copies	Food item, total	In-house method
div	8	26.07.19	negative	positive	negative	negative	Ct(LOD) in qPCR = 34	LOD is known in copies of the soybean-specific lectin gene; Ct(LOD) in qPCR is used as cut-off value	other - EURL-GMFF official method of analysis of 40-3-2 soybean, part lectin gene
div	15		negative	positive	positive	negative		Food item, total	Selection PCR-Methods
div	20		neg	pos	pos	neg	20		in-house method
div	21	12.07.19	negative	positive	positive	negative	4	Allergen-DNA	Selection PCR-Methods

Other details to the Methods

Meth. Abr.	Evaluation number	Method-No. / Test-Kit No.	Specifity	Remarks to the Method (Extraction and Determination)	Further Remarks
		Article-No. / ASU-No.	Target-DNA	e.g. Extraction / Enzymes / Clean-Up / Real Time PCR / Gel electrophoresis / Cycles	
ASU	11	L08.00-59	Lectin-F, Lectin-R	CTAB w ith/w ithout precipitation, Dneasy Mericon Food	
ASU	32	L08.00-59/2013-01	Lectin	CTAB, Proteinase K/60°C/ Clean up: Dneasy Mericon Food Kit (Qiagen)	
MS	4	1200		DNA extraction w ith proteinase K, Clean Up w ith chloroform and columns /Amplif m RealTIme PCR 45 cycles	AIIAIIA
MS	16	MS		Wizard Promega	
SFA	9	S3601		SureFood® PREP Advanced	
SFA	25				
SFA	28				
div	7		Lectin	Real Time PCR, 45 cycles	
div	8	Protocol CRLVL08/05VP Corrected version 1 dd. 20/1/2009	74 bp fragment of the soybean-specific lectin gene	NucleoSpin Food DNA extraction kit (Machery-Nagel); qPCR follow ing EURL-GMFF method CRLVL08/05VP w ith 45 cycles on a LC480 real-time qPCR machine	Sample 3: a Ct value is measured, how ever > Ct(LOD) w hich is 34; w hich means no signal >LOD for lectin qPCR
div	15	ISO 21570	81 pb	Extraction CTAB; real time PCR, 45 cycles	lectin
div	20				Limit of detection given as µg of DNA per kg of sample
div	21	Internal method		CTAB / Proteinase K / Promega Wizard DNA-CleanUp / Real-time PCR / 45 cycles	

5.2 Homogeneity

5.2.1 Mixture homogeneity before bottling

Microtracer Homogeneity Test
DLA 12-2019 Sample 1

Weight whole sample	1,00	kg
Microtracer	FSS-rot lake	
Particle size	75 – 300	µm
Weight per particle	2,0	µg
Addition of tracer	29,4	mg/kg

Result of analysis

Sample	Weight [g]	Particle number	Particles [mg/kg]
1	4,27	63	29,5
2	5,15	85	33,0
3	5,55	91	32,8
4	4,97	71	28,6
5	5,18	80	30,9
6	5,12	63	24,6
7	4,99	82	32,9
8	5,14	88	34,2

Poisson distribution		
Number of samples	8	
Degree of freedom	7	
Mean	77,7	Particles
Standard deviation	7,97	Particles
χ^2 (CHI-Quadrat)	5,73	
Probability	**57**	%
Recovery rate	105	%

Normal distribution		
Number of samples	8	
Mean	30,8	mg/kg
Standard deviation	3,16	mg/kg
rel. Standard deviaton	10,3	%
Horwitz standard deviation	9,6	%
HorRat-value	**1,1**	
Recovery rate	105	%

Microtracer Homogeneity Test
DLA 12-2019 Sample 2

Weight whole sample	1,00	kg
Microtracer	FSS-rot lake	
Particle size	75 – 300	µm
Weight per particle	2,0	µg
Addition of tracer	33,5	mg/kg

Result of analysis

Sample	Weight [g]	Particle number	Particles [mg/kg]
1	5,05	122	48,3
2	4,95	125	50,5
3	5,06	129	51,0
4	4,94	113	45,7
5	5,11	135	52,8
6	5,05	121	47,9
7	5,25	123	46,9
8	5,07	122	48,1

Poisson distribution		
Number of samples	8	
Degree of freedom	7	
Mean	123,7	Particles
Standard deviation	5,93	Particles
χ^2 (CHI-Quadrat)	1,99	
Probability	**96**	%
Recovery rate	146	%

Normal distribution		
Number of samples	8	
Mean	48,9	mg/kg
Standard deviation	2,34	mg/kg
rel. Standard deviaton	4,79	%
Horwitz standard deviation	8,91	%
HorRat-value	**0,54**	
Recovery rate	146	%

Microtracer Homogeneity Test
DLA 12-2019 Sample 3

Weight whole sample	1,00	kg
Microtracer	FSS-rot lake	
Particle size	75 – 300	µm
Weight per particle	2,0	µg
Addition of tracer	31,2	mg/kg

Result of analysis

Sample	Weight [g]	Particle number	Particles [mg/kg]
1	5,00	101	40,4
2	4,93	98	39,8
3	5,05	98	38,8
4	5,02	87	34,7
5	5,05	102	40,4
6	5,15	90	35,0
7	4,99	93	37,3
8	5,24	96	36,6

Poisson distribution		
Number of samples	8	
Degree of freedom	7	
Mean	95,7	Particles
Standard deviation	5,88	Particles
χ^2 (CHI-Quadrat)	2,53	
Probability	**92**	%
Recovery rate	121	%

Normal distribution		
Number of samples	8	
Mean	37,9	mg/kg
Standard deviation	2,33	mg/kg
rel. Standard deviaton	6,15	%
Horwitz standard deviation	9,26	%
HorRat-value	**0,66**	
Recovery rate	121	%

Microtracer Homogeneity Test
DLA 12-2019 Sample 4

Weight whole sample	1,00	kg
Microtracer	FSS-rot lake	
Particle size	75 – 300	µm
Weight per particle	2,0	µg
Addition of tracer	28,2	mg/kg

Result of analysis

Sample	Weight [g]	Particle number	Particles [mg/kg]
1	5,13	99	38,6
2	5,07	76	30,0
3	5,03	74	29,4
4	5,06	91	36,0
5	4,92	77	31,3
6	5,14	76	29,6
7	5,03	84	33,4
8	4,96	82	33,1

Poisson distribution		
Number of samples	8	
Degree of freedom	7	
Mean	82,4	Particles
Standard deviation	8,30	Particles
χ^2 (CHI-Quadrat)	5,86	
Probability	**56**	%
Recovery rate	116	%

Normal distribution		
Number of samples	8	
Mean	32,7	mg/kg
Standard deviation	3,29	mg/kg
rel. Standard deviaton	10,1	%
Horwitz standard deviation	9,5	%
HorRat-value	**1,1**	
Recovery rate	116	%

5.3 Information on the Proficiency Test (PT)

Before the PT the participants received the following information in the sample cover letter:

PT number	***DLA 12-2019***
PT name	***Allergen-Screening II - 4 Samples qualitative: Crustaceae, Egg, Fish, Milk, Molluscs, Mustard (yellow/white, brown and black), Soybean***
Sample matrix	*Samples 1-4:* *Carrier matrix / ingredients: potato powder (appr. 75%), maltodextrin (appr. 25%), other food additives and allergenic foods*
Number of samples and sample amount	*4 different Samples 1-4: 20 g each*
Storage	*Samples A + B: room temperature (long term cooled 2 - 10°C)*
Intentional use	*Laboratory use only (quality control samples)*
Parameter	*Qualitative: Crustaceae, Egg, Fish, Milk, Molluscs, Mustard (yellow/white, brown and black), Soybean (protein / DNA)* *Samples 1-4: appr. 25 - 250 mg/kg*
Methods of analysis	*The analytical methods ELISA (+ Lateral Flow) and PCR can be applied for qualitative determinations.*
Notes to analysis	*The analysis of PT samples should be performed like a routine laboratory analysis.* *In general we recommend to homogenize a representative sample amount before analysis according to good laboratory practice, especially in case of low sample weights.*
Result sheet	*One result each should be determined for Samples 1-4.* *The results should be filled in the result submission file.*
Units	*posititv / negativ (limit of detection mg/kg)*
Number of digits	*at least 2*
Result submission	*The result submission file should be sent by e-mail to:* ***pt@dla-lvu.de***
Deadline	***the latest 30th August 2019***
Evaluation report	*The evaluation report is expected to be completed 6 weeks after deadline of result submission and sent as PDF file by e-mail.*
Coordinator and contact person of PT	*Matthias Besler-Scharf PhD*

* Control of mixture homogeneity and qualitative testings are carried out by DLA. Any testing of the content, homogeneity and stability of PT parameters is subcontracted by DLA.

6. Index of participant laboratories

Teilnehmer / Participant	Ort / Town	Land / Country
		SPAIN
		GREAT BRITAIN
		SPAIN
		USA
		SPAIN
		CANADA
		CANADA
		ITALY
		Germany
		FRANCE
		Germany
		ITALY
		HUNGARY
		GREAT BRITAIN
		CANADA
		BELGIUM
		Germany
		SPAIN
		SWITZERLAND
		SPAIN
		ITALY
		BELGIUM
		Germany
		Germany
		Germany
		Germany
		AUSTRIA
		GREAT BRITAIN
		Germany
		FRANCE
		GREAT BRITAIN
		GREAT BRITAIN
		SLOVAKIA
		Germany
		GREAT BRITAIN

[Die Adressdaten der Teilnehmer wurden für die allgemeine Veröffentlichung des Auswerte-Berichts nicht angegeben.]

[The address data of the participants were deleted for publication of the evaluation report.]

7. Index of references

1. DIN EN ISO/IEC 17025:2005; Allgemeine Anforderungen an die Kompetenz von Prüf- und Kalibrierlaboratorien / General requirements for the competence of testing and calibration laboratories
2. DIN EN ISO/IEC 17043:2010; Konformitätsbewertung – Allgemeine Anforderungen an Eignungsprüfungen / Conformity assessment – General requirements for proficiency testing
3. ISO 13528:2015 & DIN ISO 13528:2009; Statistische Verfahren für Eignungsprüfungen durch Ringversuche / Statistical methods for use in proficiency testing by inter-laboratory comparisons
4. ASU §64 LFGB: Planung und statistische Auswertung von Ringversuchen zur Methoden-validierung / DIN ISO 5725 series part 1, 2 and 6 Accuracy (trueness and preci-sion) of measurement methods and results
5. Verordnung / Regulation 882/2004/EU; Verordnung über über amtliche Kontrollen zur Überprüfung der Einhaltung des Lebensmittel- und Futtermittelrechts sowie der Bestimmungen über Tiergesundheit und Tierschutz / Regulation on official controls performed to ensure the verification of compliance with feed and food law, animal health and animal welfare rules
6. Evaluation of analytical methods used for regulation of food and drugs; W. Hor-witz; Analytical Chemistry, 54, 67-76 (1982)
7. The International Harmonised Protocol for the Proficiency Testing of Ananlytical Laboratories ; J.AOAC Int., 76(4), 926 – 940 (1993)
8. A Horwitz-like funktion describes precision in proficiency test; M. Thompson, P.J. Lowthian; Analyst, 120, 271-272 (1995)
9. Protocol for the design, conduct and interpretation of method performance studies; W. Horwitz; Pure & Applied Chemistry, 67, 331-343 (1995)
10. Recent trends in inter-laboratory precision at ppb and sub-ppb concentrations in relation to fitness for purpose criteria in proficiency testing; M. Thompson; Ana-lyst, 125, 385-386 (2000)
11. The International Harmonised Protocol for the Proficiency Testing of Analytical Chemistry Laboratories; Pure Appl Chem, 78, 145 – 196 (2006)
12. AMC Kernel Density - Representing data distributions with kernel density estim-ates, amc technical brief, Editor M Thompson, Analytical Methods Committee, AMCTB No 4, Revised March 2006 and Excel Add-in Kernel.xla 1.0e by Royal Society of Chemistry
13. EURACHEM/CITAC Leitfaden, Ermittlung der Messunsicherheit bei analytischen Messun-gen (2003); Quantifying Uncertainty in Analytical Measurement (1999)
14. GMP+ Feed Certification scheme, Module: Feed Safety Assurance, chapter 5.7 Check-ing procedure for the process accuracy of compound feed with micro tracers in GMP+ BA2 Control of residues, Version: 1st of January 2015 GMP+ International B.V.
15. MTSE SOP No. 010.01 (2014): Quantitative measurement of mixing uniformity and carry-over in powder mixtures with the rotary detector technique, MTSE Micro Tracers Services Europe GmbH
16. Homogeneity and stability of reference materials; Linsinger et al.; Accred Qual Assur, 6, 20-25 (2001)
17. AOAC Official Methods of Analysis: Guidelines for Standard Method Performance Re-quirements, Appendix F, p. 2, AOAC Int (2016)
18. Codex Alimentarius Commission (2010) - Guidelines on performance criteria and validation of methods for detection, identification and quantification of specific DNA sequences and specific protiens in foods, CAC/GL 74-2010
19. DIN EN ISO 15633-1:2009; Nachweis von Lebensmittelallergenen mit immunologischen Verfahren - Teil 1: Allgemeine Betrachtungen / Foodstuffs - Detection of food allergens by immunological methods - Part 1: General considerations
20. DIN EN ISO 15634-1:2009; Nachweis von Lebensmittelallergenen mit molekularbiologischen Verfahren - Teil 1: Allgemeine Betrachtungen / Foodstuffs - Detection of food allergens by molecular biological methods - Part 1: General considerations
21. DIN EN ISO 15842:2010 Lebensmittel – Nachweis von Lebensmittelallergenen – Allgemeine Betrachtungen und Validierung von Verfahren / Foodstuffs -

Detection of food allergens - General considerations and validation of methods
22. Ministry of Health and Welfare, JSM, Japan 2006
23. Working Group Food Allergens, Abbott et al., Validation Procedures for Quantitative Food Allergen ELISA Methods: Community Guidance and Best Practices JAOAC Int. 93:442-50 (2010)
24. Working Group on Prolamin Analysis and Toxicity (WGPAT): Méndez et al. Report of a collaborative trial to investigate the performance of the R5 enzyme linked immunoassay to determine gliadin in gluten-free food. Eur J Gastroenterol Hepatol. 17:1053-63 (2005)
25. DLA Publikation: Performance of ELISA and PCR methods for the determination of allergens in food: an evaluation of six years of proficiency testing for soy (Glycine max L.) and wheat gluten (Triticum aestivum L.); Scharf et al.; J Agric Food Chem. 61(43):10261-72 (2013)
26. EFSA (2014) Scientific Opinion on the evaluation of allergenic foods and food ingredients for labelling purposes1, EFSA Panel on Dietetic Products, Nutrition and Allergies (NDA), European Food Safety Authority (EFSA), Parma, Italy, EFSA Journal 2014;12(11):3894
27. IRMM, Poms et al.; Inter-laboratory validation study of five different commercial ELISA test kits for determination of peanut residues in cookie and dark chocolate; European Commission, Joint Research Centre, Belgium; GE/R/FSQ/D08/05/2004
28. Jayasena et al. (2015) Comparison of six commercial ELISA kits for their specificity and sensitivity in detecting different major peanut allergens. J Agric Food Chem. 2015 Feb 18;63(6):1849-55
29. ASU §64 LFGB L 06.00-56 Bestimmung von Sojaprotein in Fleisch und Fleischerzeugnissen Enzymimmunologisches Verfahren (2007) [Determination of soyprotein in meat and meat products by enzyme immunoassay]
30. ASU §64 LFGB L 00.00-69 Bestimmung von Erdnuss-Kontaminationen in Lebensmitteln mittels ELISA im Mikrotiterplattensystem (2003) [Foodstuffs, determination of peanut contamintions in foodstuffs by ELISA in microtiterplates]
31. ASU §64 LFGB L 44.00-7 Bestimmung von Haselnuss-Kontaminationen in Schokolade und Schokoladenwaren mittels ELISA im Mikrotiterplattensystem (2006) [Foodstuffs, determination of hazelnut contamintions in chocolate and chocolate products by ELISA in microtiterplates]