

Evaluation Report proficiency test

DLA 34/2018

GMO-Screening qualitative:

5 Samples with positive/negative amounts of 35S, NOS, FMV, pNOS/nptll, 35S-Pat, Cry1Ab/Ac, CTP2-CP4 EPSPS / GMO-Maize (Bt11, MIR604) and GMO-Soya (RR GTS 40-3-2, RR2 MON89788)

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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.

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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

The test materials are 5 different mixtures of common in commerce food mixtures from European, US-American and Asian suppliers (s. table 1). The raw materials were crushed, sieved (mesh <250 μm to <600 μm), mixed and homogenized. The composition of the samples is given in table 1.

Before homogenization microtracer particles were added in order to check the accuracy of mixing. After homogenization during bottling aliquots were taken for microtracer analysis (s. 2.1.1).

After homogenisation the samples were portioned to approximately 10 g into metallised PET film bags.

Table 1: Composition of DLA-Samples

DLA- Sample	Ingredients (per 100 g)	GMO-Con- tent Maize	GMO-Con- tent Soya
1	Wheat flour Type 505 (90 g) Ingredients: Wheat Nutrients per 100 g: Protein 11 g, Carbohydrates 72 g, Fat 1,1 g	-	-
	Soya Flour, European Supplier (7,5 g) Ingredients: Soya flour toasted Nutrients per 100 g: Protein 37 g	-	-
	Soya Chunks, USA-Supplier (2,7 g) Ingredients: Soybean Flour Nutrients per 100 g: Protein 47 g, Carbohydrates 17 g, Fat 0,8 g	-	positive (GM-Soya experimental)
2	Wheat flour Type 505 (90 g) Ingredients: Wheat Nutrients per 100 g: Protein 11 g, Carbohydrates 72 g, Fat 1,1 g	-	-
	Soya Flour, Chinese Supplier (10 g) Ingredients: Soya flour, not toasted Nutrients per 100 g: Protein 41 g	-	-
_		-	-
3	Wheat flour Type 505 (90 g) Ingredients: Wheat Nutrients per 100 g: Protein 11 g, Carbohydrates 72 g, Fat 1,1 g	-	-
	Maize Semolina, European-Supplier (10 g) Ingredients: Maize Flour Nutrients per 100 g:	-	-
	Protein 7,5 g, Carbohydrates 74 g, Fat 1 g Soya Flour, European Supplier (10 g) Ingredients: Soya flour toasted Nutrients per 100 g: Protein 37 g	-	-
4	Wheat flour Type 505 (90 g) Ingredients: Wheat Nutrients per 100 g: Protein 11 g, Carbohydrates 72 g, Fat 1,1 g	-	-
5	Wheat flour Type 505 (90 g) Ingredients: Wheat Nutrients per 100 g: Protein 11 g, Carbohydrates 72 g, Fat 1,1 g	-	-
	Maize Semolina, European-Supplier (13 g) Ingredients: Maize Flour Nutrients per 100 g:	-	-
	Protein 7,5 g, Carbohydrates 74 g, Fat 1 g Maize Flour, USA-Supplier (6,6 g) Ingredients: Maize Flour Nutrients per 100 g: Protein 9 g, Carbohydrates 79 g, Fat 0 g	positive (GM-Maize experimental)	-

 ${\it Note:}$ The metrological traceability of temperature, mass and volume during production of the PT samples is ensured by DAkkS calibrated reference materials.

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 \geq 5 % is equivalent to a good homogeneous mixture and of \geq 25% to an excellent mixture [14, 15].

The microtracer analysis of the present PT samples 1, 2, 3 and 5 showed probabilities of 100%, 20%, 99% 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) [16, 17]. This gave HorRat values of 0,39, 1,6, 0,60 and 1,2, respectively. The HorRat value of sample 3 was slightly increased, while the probability was well > 25%. The results of microtracer analysis are given in the documentation. The PT sample 4 was not tested by microtracer analysis, because it contained neither soy nor maize products.

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. 0,48 (23,5°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 5) were sent to every participating laboratory in the 22^{nd} week of 2018. The testing method was optional. The tests should be finished at July 13^{th} 2018 the latest.

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

DLA 34-2018 - GMO-Screening qualitative: 5 Samples with positive/negative amounts of Screening Targets 35S, NOS, FMV, CTP2-CP4 EPSPS / GMO-Maize (Bt11, MIR604) and GMO-Soya (RR GTS 40-3-2, RR2 MON89788)

There are 5 different test samples wich possibly contain the mentioned parameters. The indication of results and evaluation will be done exclusively qualitative (positive/negative). Results for specific sequences, screening sequences and other events can be analyzed.

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 or were available on our website. 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.

All 27 participants submitted their results in time.

3. Evaluation

The evaluation of the GMO-screening proficiency test was done exclusively qualitative.

The results are presented for all 5 test samples in separate tables for each parameter p-35S, t-NOS, FMN, p-NOS/nptII, CTP2-CP4 EPSPS, 35S-Pat, Cry1Ab/Ac, GMO-Maize Bt11, GMO-Maize MIR604, Maize-DNA and GMO-Soya RR (GTS 40-3-2), GMO-Soya RR2 (MON89788), Lectin-DNA and other DNA.

3.1 Agreement with consensus values from participants

The qualitative evaluation of the 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 $\geq 75\%$ 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 results of each participant was based on the agreement of the indicated results (positive or negative) with the **spiking of the five PT-samples**. A consensus value is determined unless \geq 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 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 participant results and evaluation are tabulated as follows:

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
	pos/neg	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	Sample 5
Number positive					
Number negative					
Percent positive					
Percent negative					
Consensus value					
Spiking					

4.1 Proficiency Test

4.1.1 Results: 35S-Screening-Sequence

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
35S	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples	
1	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
2	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
3	positive	negative	positive	positive	positive	3/5 (60%)	3/5 (60%)	
4	positive	positive	negative	negative	positive	4/5 (80%)	4/5 (80%)	
5	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
6	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
7	positive	negative	negative	positive	positive	4/5 (80%)	4/5 (80%)	
8	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
9	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
10a	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
10b	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
11	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
12	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
13	positive	positive	negative	negative	positive	4/5 (80%)	4/5 (80%)	
14	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
15	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
16	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
17	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
18	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
19	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
20	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
21	positive	positive	positive		positive	2/4 (50%)	2/4 (50%)	Sample 4: +/-
22	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
23	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
24	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
25	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	Samples 2,3,4 traces (< 0,1%)
26	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
27	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Number positive	28	3	2	2	28
Number negative	0	25	26	25	0
Percent positive	100	11	7	7	100
Percent negative	0	89	93	93	0
Consensus value	positive	negative	negative	negative	positive
Spiking	positive	negative	negative	negative	positive

<u>Comments on results:</u>

For all 5 samples consensus values with two times 100% and 93% each and 89% positive or negative results were obtained, respectively. The consensus values are in agreement with the addition of the GMO-containing ingredients (spiking).

<u>4.1.2 Results:</u> NOS-Screening-Sequence

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
NOS	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples	
1	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
2	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
3	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
4	positive	positive	negative	negative	positive	4/5 (80%)	4/5 (80%)	
5	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
6	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
7	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
8	negative	negative	negative	negative	negative	3/5 (60%)	3/5 (60%)	
9	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
10a	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
10b	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
11	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
12	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
13	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
14	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
15	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
16	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
17	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
18	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
19	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
20	-	-	-	-	-			
21	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
22	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
23	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
24	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
25	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
26	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
27	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Number positive	26	1	0	0	26
Number negative	1	26	27	27	1
Percent positive	96	4	0	0	96
Percent negative	4	96	100	100	4
Consensus value	positive	negative	negative	negative	positive
Spiking	positive	negative	negative	negative	positive

<u>Comments on results:</u>

For all 5 samples consensus values with two times 100% and three times 96%, respectively.

The consensus values are in agreement with the addition of the ${\tt GMO-containing}$ ingredients (spiking).

4.1.3 Results: FMV-Screening-Sequence

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
FMV	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples	
1	-	-	-	-	-			
2	negative	negative	negative	negative	negative	4/4 (100%)	4/5 (80%)	
3	positive	negative	negative	negative	negative	4/4 (100%)	5/5 (100%)	
4	-	-	-	-	-			
5	negative	negative	negative	negative	negative	4/4 (100%)	4/5 (80%)	Sample 1: FMV positive <0,9%
6	positive	negative	negative	negative	negative	4/4 (100%)	5/5 (100%)	
7	-	-	-	-	-			
8	negative	negative	negative	negative	negative	4/4 (100%)	4/5 (80%)	
9	negative	positive	negative	negative	positive	2/4 (50%)	2/5 (40%)	
10	positive	negative	negative	negative	negative	4/4 (100%)	5/5 (100%)	
11	positive	negative	negative	negative	negative	4/4 (100%)	5/5 (100%)	
12	positive	negative	negative	negative	-	3/3 (100%)	4/4 (100%)	
13	positive	negative	negative	negative	negative	4/4 (100%)	5/5 (100%)	
14	negative	negative	negative	negative	negative	4/4 (100%)	4/5 (80%)	
15	positive	negative	negative	negative	negative	4/4 (100%)	5/5 (100%)	
16	positive	negative	negative	negative	negative	4/4 (100%)	5/5 (100%)	
17	negative	negative	negative	negative	negative	4/4 (100%)	4/5 (80%)	
18	positive	negative	negative	negative	negative	4/4 (100%)	5/5 (100%)	
19	negative	negative	negative	negative	negative	4/4 (100%)	4/5 (80%)	
20	-	-	-	-	-			
21	positive	negative	negative	negative	negative	4/4 (100%)	5/5 (100%)	
22	positive	negative	negative	negative	negative	4/4 (100%)	5/5 (100%)	
23	positive	negative	negative	negative	negative	4/4 (100%)	5/5 (100%)	
24	positive	negative	negative	negative	negative	4/4 (100%)	5/5 (100%)	
25	positive	negative	negative	negative	negative	4/4 (100%)	5/5 (100%)	
26	negative	negative	negative	negative	negative	4/4 (100%)	4/5 (80%)	
27	negative	negative	negative	negative	negative	4/4 (100%)	4/5 (80%)	

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Number positive	14	1	0	0	1
Number negative	9	22	23	23	21
Percent positive	61	4	0	0	5
Percent negative	39	96	100	100	95
Consensus value	keiner	negative	negative	negative	negative
Spiking	positive	negative	negative	negative	negative

<u>Comments on results:</u>

For samples 2-5 consensus values with two times 100%, 96% and 95% positive or negative results were obtained, respectively. The consensus values are in agreement with the addition of the GMO-containing ingredients (spiking).

For sample 1 no consensus with $\geq 75\%$ positive or negative results was obtained.

4.1.4 Results: p-NOS/nptII-Screening-Sequence

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
p-NOS/nptII	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples	
1	-	-	-	-	-			
2	-	-	-	-	-			
3	negative	negative	negative	negative	positive			
4	-	-	-	-	-			
5	-	-	-	-	-			
6	negative	negative	negative	negative	positive			
7	-	-	-	-	-			
8	-	-	-	-	-			
9	-	-	-	-	-			
10	-	-	-	-	-			
11	negative	negative	negative	negative	negative			
12	-	negative	negative	negative	-			
13	-	-	-	-	-			
14	neg / pos	negative	negative	negative	positive			Sample 1 p-NOS neg, nptll pos
15	negative	negative	negative	negative	negative			
16	-	-	-	-	-			
17	-	-	-	-	-			
18	-	-	-	-	-			
19	-	-	-	-	-			
20	-	-	-	-	-			
21	negative	negative	negative	negative	negative			
22	-	-	-	-	-			
23	negative	negative	negative	negative	negative			
24	-	-	-	-	-			
25	-	-	-	-	-			
26	-	-	-	-	-			
27	-	-	-	_	-			

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	
Number positive	0	0	0	0	3	
Number negative	6	8	8	8	4	
Percent positive	0	0	0	0	43	
Percent negative	100	100	100	100	57	
Consensus value	negative	negative	negative	negative	none	
Spiking	negative	negative	negative	negative	positive *	

^{*} nptll positive / p-NOS unkown

<u>Comments on results:</u>

The participants applied methods with different targets. On one hand for the detection of the constructs p-NOS-nptII (participants 11, 15 and 23) and on the other for separate detection of nptII (participant 6 and 14) and p-NOS (participant 14).

In sample 5, the separate detection of nptII was possible, while the construct p-NOS-nptII was not detectable.

Due to the lack of data for the different targets, no evaluation of the results was made.

4.1.5 Results: CTP2-CP4 EPSPS-Screening-Sequence

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
CTP2-CP4 EPSPS	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples	
1	-	-	=	-	-			
2	-	-	-	-	-			
3	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
4	-	-	-	-	-			
5	-	-	-	-	-			
6	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
7	-	-	-	-	-			
8	-	-	-	-	-			
9	-	-	-	-	-			
10	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
11	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
12	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
13	-	-	-	-	-			
14	negative	negative	negative	negative	negative	3/5 (100%)	3/5 (100%)	
15	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
16	-	-	=	-	-			
17	-	-	-	-	-			
18	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
19	-	-	-	-	-			
20	-	-	-	-	-			
21	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
22	negative	negative	negative	negative	negative	3/5 (100%)	3/5 (100%)	
23	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
24	-	-	=	-	-			
25	_		-	_	-			
26	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
27	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Number positive	11	0	0	0	11
Number negative	2	13	13	13	2
Percent positive	85	0	0	0	85
Percent negative	15	100	100	100	15
Consensus value	positive	negative	negative	negative	positive
Spiking	positive	negative	negative	negative	positive

<u>Comments on results:</u>

For all 5 samples consensus values with three times 100% and two times 85% positive or negative results were obtained.

The consensus values are in agreement with the addition of the ${\tt GMO-containing}$ ingredients (spiking).

4.1.6 Results: 35S-Pat-Screening-Sequence

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
35S-Pat	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples	
1	-	-	-	-	-			
2	-	-	-	-	-			
3	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
4	-	-	-	-	-			
5	-	-	-	-	-			
6	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
7	-	-	-	-	-			
8	-	-	-	-	-			
9	-	-	-	-	-			
10	-	-	-	-	-			
11	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
12	-	negative	negative	negative	positive	4/4 (100%)	4/4 (100%)	
13	-	-	-	-	-			
14	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
15	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
16	-	-	-	-	-			
17	negative	negative	positive	positive	negative	1/5 (20%)	1/5 (20%)	
18	-	-	-	-	-			
19	-	-	-	-	-			
20	-	-	-	-	-			
21	-	negative	negative	negative	positive	4/4 (100%)	4/4 (100%)	
22	-	-	-	-	-			
23	positive	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
24	-	-	-	-	-			
25	-	-	-	-	-			
26	-	-	-	-	-			
27	-	-	-	-	-			

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Number positive	6	0	1	1	8
Number negative	lumber negative 1		8	8	1
Percent positive	86	0	11	11	89
Percent negative	14	100	89	89	11
Consensus value	us value positive r		negative	negative	positive
Spiking	positive	negative	negative	negative	positive

<u>Comments on results:</u>

For all 5 samples consensus values with three times 89% and one time 100% and 86% each positive or negative results were obtained. The consensus values are in agreement with the addition of the GMO-containing ingredients (spiking).

4.1.7 Results: Cry1Ab/Ac-Screening-Sequence

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
Cry1Ab/Ac	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples	
1	-	-	-	-	-			
2	-	-	-	-	-			
3	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
4	-	-	-	-	-			
5	_	-	-	-	-			
6	-	-	-	-	-			
7	-	-	-	-	-			
8	-	-	-	-	-			
9	-	-	-	-	-			
10	-	-	-	-	-			
11	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
12	-	negative	negative	negative	-	3/3 (100%)	3/3 (100%)	
13	-	-	-	-	-			
14	_	-	-	-	-			
15	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
16	-	-	-	-	-			
17	-	-	-	-	-			
18	-	-	-	-	-			
19	-	-	-	-	-			
20	-	-	-	-	negative	1/1 (100%)	1/1 (100%)	
21	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
22	-	-	-	-	-			
23	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
24	-	-	-	-	-			
25	-	-	-	-	-			
26	-	-	-	-	-			
27	-	-	-	-	-			

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

<u>Comments on results:</u>

For all 5 samples consensus values with four times 100% and one time 83% positive or negative results were obtained.

The consensus values are in agreement with the addition of the GMO-containing ingredients (spiking).

4.1.8 Results: GMO-Maize Bt11

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
GMO-Maize (bt11)	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples	
1	positive	positive	negative	negative	positive	3/5 (80%)	3/5 (80%)	
2	-	-	-	-	-			
3	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
4	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
5	-	-	-	-	-			
6	-	-	-	-	-			
7	-	-	-	-	-			
8	-	-	-	-	-			
9	-	-	-	-	-			
10	-	-	-	-	-			
11	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
12	-	negative	negative	negative	-	3/3 (100%)	3/3 (100%)	
13	-	-	-	-	-			
14		-	-	-	-			
15	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
16	-	-	-	-	-			
17	-	-	-	-	-			
18	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
19	-	-	-	-	-			
20	-	-	-	-	-			
21	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
22	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
23	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
24	-	-	=	-	-			
25	-	-	=	-	-			
26	negative	negative	negative	negative	negative	4/5 (80%)	4/5 (80%)	
27	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Number positive	1	1	0	0	10
Number negative	10	11	12	12	1
Percent positive	9	8	0	0	91
Percent negative	91	92	100	100	9
Consensus value	negative	negative	negative	negative	positive
Spiking	negative	negative	negative	negative	positive

<u>Comments on results:</u>

For all 5 samples consensus values with two times 100% and 91% and one time 92% positive or negative results were obtained.

The consensus values are in agreement with the addition of the ${\sf GMO-containing}$ ingredients (spiking).

4.1.9 Results: GMO-Maize MIR604

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
GMO-Maize (MIR604)	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples	
1	-	-	-	-	-			
2	-	-	-	-	-			
3	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
4	-	-	-	-	-			
5	-	-	-	-	-			
6	-	-	-	-	-			
7	-	-			-			
8	-	-	-	-	-			
9	-	-	-	-	-			
10	-	-	-	-	-			
11	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
12	_	negative	negative	negative	-	3/3 (100%)	3/3 (100%)	
13	-	-	-	-	-			
14	-	-	-	-	-			
15	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
16	-	-	-	-	-			
17	-	-	-	-	-			
18	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
19	-	-	-	-	-			
20	-	-	-	-	-			
21	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
22	-	-	-	-	-			
23	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
24	-	-	-	-	-			
25	-	-	-	-	-			
26	negative	negative	negative	negative	positive	5/5 (100%)	5/5 (100%)	
27	negative	negative	negative	negative	negative	4/5 (80%)	4/5 (80%)	

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Number positive	0	0	0	0	7
Number negative	8	9	9	9	1
Percent positive	0	0	0	0	88
Percent negative	100	100	100	100	13
Consensus value	negative	negative	negative	negative	positive
Spiking	negative	negative	negative	negative	positive

<u>Comments on results:</u>

For all 5 samples consensus values with four times 100% and one time 88% positive or negative results were obtained.

The consensus values are in agreement with the addition of the GMO-containing ingredients (spiking).

4.1.10 Results: Maize-DNA (Maize-specific)

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
Maize	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples	
1	positive	negative	positive	negative	positive	4/4 (100%)	4/5 (80%)	
2	-	-	-	-	-			
3	positive	negative	positive	negative	positive	4/4 (100%)	4/5 (80%)	
4	negative	negative	positive	negative	positive	4/4 (100%)	5/5 (100%)	
5	-	-	-	-	-			
6	-	-	-	-	-			
7	positive	negative	positive	negative	positive	4/4 (100%)	4/5 (80%)	
8	-	-	-	-	-			
9	-	-	-	-	-			
10	-	-	-	-	-			
11	positive	negative	positive	negative	positive	4/4 (100%)	4/5 (80%)	
12	-	negative	negative	negative	-	2/3 (66%)	2/3 (66%)	
13	-	-	_	-	-			
14	-	-	-	-	-			
15	positive	negative	positive	negative	positive	4/4 (100%)	4/5 (80%)	
16	-	-	-	-	-			
17	-	-	-	-	-			
18	positive	negative	positive	negative	positive	4/4 (100%)	4/5 (80%)	
19	-	-	-	-	-			
20	-	-	-	-	-			
21	negative	negative	positive	negative	positive	4/4 (100%)	5/5 (100%)	
22	negative	negative	positive	negative	positive	4/4 (100%)	5/5 (100%)	
23	-	-	-	-	-			
24	-	-	-	-	-			
25	-	-	-	-	-			
26	negative	negative	positive	negative	positive	4/4 (100%)	5/5 (100%)	
27	negative	negative	positive	negative	positive	4/4 (100%)	5/5 (100%)	

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Number positive	6	0	11	0	11
Number negative	5	12	1	12	0
Percent positive	55	0	92	0	100
Percent negative	45	100	8	100	0
Consensus value	none	negative	positive	negative	positive
Spiking	negative	negative	positive	negative	positive

<u>Comments on results:</u>

For samples 2-5 consensus values with three times 100% and one time 92% positive or negative results were obtained, respectively. The consensus values are in agreement with the addition of maize-containing ingredients (spiking).

For sample 1 no consensus with $\geq 75\%$ positive or negative results was obtained.

4.1.11 Results: GMO-Soya RR (GTS 40-3-2)

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
GM-Soya RR (GTS 40-3-2)	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples	
1	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
2	-	-	-	-	-			
3	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
4	positive	positive	negative	negative	negative	4/5 (80%)	4/5 (80%)	
5	-	-	-	-	-			
6	_	-	-	-	-			
7	-	-	-	-	-			
8	-	-	-	-	-			
9	-	-	-	-	-			
10a	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
10b	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
11	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
12	positive	negative	negative	negative	-	4/4 (100%)	4/4 (100%)	
13	-	-	-	-	-			
14	_	-	-	-	-			
15	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
16	-	-	-	-	-			
17	-	-	-	-	-			
18	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
19	-	-	-	-	-			
20	positive	-	-	-	negative	2/2 (100%)	2/2 (100%)	
21	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
22	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
23	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
24	-	-	-	-	-			
25	-	-	-	-	-			
26	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
27	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Number positive	15	1	0	0	0
Number negative	0	13	14	14	14
Percent positive	100	7	0	0	0
Percent negative	0	93	100	100	100
Consensus value	positive	negative	negative	negative	negative
Spiking	positive	negative	negative	negative	negative

<u>Comments on results:</u>

For all 5 samples consensus values with four times 100% and one time 93% positive or negative results were obtained, respectively. The consensus values are in agreement with the addition of the GMO-containing ingredients (spiking).

4.1.12 Results: GMO-Soya RR2 (MON89788)

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
GM-Soya RR2 (MON89788)	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples	
1	-	-	-	-	-			
2	-	-	-	-	-			
3	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
4	positive	positive	negative	negative	negative	4/5 (80%)	4/5 (80%)	
5	-	-	-	-	-			
6	-	-	-	-	-			
7	-	-	-	-	-			
8	-	-	-	-	-			
9	-	-	-	-	-			
10a	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
10b	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
11	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
12	positive	negative	negative	negative	-	4/4 (100%)	4/4 (100%)	
13	-	-	-	-	-			
14	-	-	-	-	-			
15	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
16	-	-	-	-	-			
17	-	-	=	=	-			
18	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
19	-	-	-	-	-			
20	-	-	-	-	-			
21	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
22	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
23	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	
24	-	-	-	-	-			
25	-	-	-	-	-			
26	negative	negative	negative	negative	negative	4/5 (80%)	4/5 (80%)	
27	positive	negative	negative	negative	negative	5/5 (100%)	5/5 (100%)	

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Number positive	12	1	0	0	0
Number negative	1	12	13	13	12
Percent positive	92	8	0	0	0
Percent negative	8	92	100	100	100
Consensus value	positive	negative	negative	negative	negative
Spiking	positive	negative	negative	negative	negative

<u>Comments on results:</u>

For all 5 samples consensus values with three times 100% and two times 92% positive or negative results were obtained, respectively. The consensus values are in agreement with the addition of the GMO-containing ingredients (spiking).

4.1.13 Results: Lectin-DNA (Soya-specific)

Evaluation number	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Qualitative Valuation	Qualitative Valuation	Remarks
Lectin DNA	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	Agreement with consensus value	Agreement with spiking of samples	
1	positive	positive	positive	positive	negative	4/4 (100%)	4/5 (80%)	
2	-	-	-	-	-			
3	positive	positive	positive	positive	positive	4/4 (100%)	3/5 (60%)	
4	positive	positive	positive	negative	negative	3/4 (75%)	5/5 (100%)	
5	-	-	-	-	-			
6	-	-	-	-	-			
7	-	-	-	-	-			
8	-	-	-	-	-			
9	-	-	-	-	-			
10	-	-	-	-	-			
11	positive	positive	positive	positive	positive	4/4 (100%)	3/5 (60%)	
12	-	negative	negative	negative	-	0/3 (0%)	1/3 (33%)	
13	-	-	-	-	-			
14	-	-	-	-	-			
15	positive	positive	positive	positive	positive	4/4 (100%)	3/5 (60%)	
16	-	-	-	-	-			
17	-	-	=	-	-			
18	positive	positive	positive	positive	positive	4/4 (100%)	3/5 (60%)	
19	-	-	-	-	-			
20	positive	positive	positive	positive	positive	4/4 (100%)	3/5 (60%)	
21	positive	positive	positive	positive	positive	4/4 (100%)	3/5 (60%)	
22	positive	positive	positive	positive	positive	4/4 (100%)	3/5 (60%)	
23				-				
24	-	-	-	-	-			
25	-	-	-	-	-			
26	positive	positive	positive	negative	negative	3/4 (75%)	5/5 (100%)	
27	positive	positive	positive	positive	positive	4/4 (100%)	3/5 (60%)	

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Number positive	11	11	11	9	8
Number negative	0	1	1	3	3
Percent positive	100	92	92	75	73
Percent negative	0	8	8	25	27
Consensus value	positive	positive	positive	positive	keiner
Spiking	positive	positive	positive	negative	negative

Comments on results:

For samples 1-4 consensus values with one time 100%, two times 92% and one time 75% positive or negative results were obtained, respectively. For samples 1-3 the consensus values are in agreement with the addition of soya-containing ingredients (spiking). Soya was not added to samples 4 and 5, however, traces of soya can not be excluded.

For sample 5 no consensus with $\geq 75\%$ positive or negative results was obtained.

4.1.14 Results: Other Parameters (DNA)

Evaluation number	Parameter	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Remarks
	other DNA	pos/neg	pos/neg	pos/neg	pos/neg	pos/neg	
3a	NK603	negative	negative	negative	negative	positive	
3b	CaMV	negative	negative	negative	negative	negative	
6	BAR	negative	negative	negative	negative	negative	
10a	Plant-DNA	positive	positive	positive	positive	positive	
10b	pat	negative	negative	negative	negative	positive	
10c	bar	negative	negative	negative	negative	negative	
11a	bar	negative	negative	negative	negative	negative	
11b	p-35-nptll / MON89034	negative	negative	negative	negative	negative	
11c	TC1507 Maize	negative	negative	negative	negative	positive	
11d	A2704-12 Soja	positive	negative	negative	negative	negative	
11e	NK603 / MON810	negative	negative	negative	negative	positive	
19	bar	negative	negative	negative	negative	negative	
21a	CaMV	positive	positive / negative	positive / negative	positive	positive / negative	
21b	GMO-Maize (NK603)	-	-	-	-	positive	
21c	GVO-Mais (MON88017)	-	-	1	-	positive	
21d	Soya A5547	negative	-	-	-	-	
21e	Soya A2704	positive / negative	-	-	-	-	
22a	T-35S	negative	negative	negative	negative	negative	
22b	bar	negative	negative	negative	negative	negative	
22c	PAT	positive	negative	negative	negative	negative	
22d	GOX	negative	negative	negative	negative	negative	
23a	T25	positive	-	1	-	-	also see documentation
23b	bar	negative	negative	negative	negative	negative	
23c	MON88017- Maize	-	-	-	-	positive	
23d	NK603-Maize	negative	-	-	-	positive	
23e	A2704-Soya	negative	-	-	-	-	
23f	A5547-Soya	negative	-	ı	-	-	
26a	Pat	-	-	ı	-	positive	
26b	NK603	-	-	-	-	positive	
26c	TC1507	-	-	-	-	positive	

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).

see following pages

5.1.1 35S-Screening Sequence

Evaluation number	Date of Analysis	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
358	Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA amount	e.g real time PCR / gel electrophoresis / cycles / amplificate length / reference material	
1	13.07.18		0,10%	In house method	Magnetic Bead Extraction	Gel Electrophoresis	
2	20.06.2018.	35S	0.01	Biotecon Screening 1 Lyokit	Biotecon Sample Prep. Kit 3	Real-time PCR	
3	08.06.18				·	Real-time PCR	
4	13.06.18	195 bp		as per ASU § 64 L00.00-118 (primer sequence, PCR premix, PCR program)	DNA-extraction by Wizard-Kit from Promega	gel electrophoresis	
5	26.06.18	p35S CAMV	LOD: 5 DNA-copies	Congen, Surefood GMO screen 35S, NOS & FMV (S2026)	Kit: Machery Nagel, Nucleospin DNA yield: >50 ng/µL A260/280: >1,6	qPCR, 45 cycles, reference material 1% CRM	Positive: >or = 0,9% Negative: <0,9%
6	11.07.18	35s	0,10%	Congen SureFood GMO 1 Kit	SureFood Prep Advanced Kit	Real-Time PCR	
7	11.07.18	target-Sequence	<0,01%	in House Method	prepman ultra applyed biosystem-no clean up- taqman universal master mix applyed biosystem	real time PCR	sample 4: ct 39,95 and 39,77
8	28.06.18			GEN-IAL		Real Time PCR	
9	18.06.18			Biotecon		RealTime PCR	
10	08.06.		10 copies	GEN-IAL GmbH	Genomic DNA from Food (Macherey-Nagel)	Real Time PCR	
10	13.06.	DNA	0,02 %	ASU §64 method 00.00 122	in house method	Real Time PCR	
11	12.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
12							
13	25.06.18	Target / DNA	0,1%	Manufacturer Bioside / ISO 21569:2005/Amd 1:2013	Nucleo spin Food Macherey-Nagel	Real Time PCR	
14	06.07.18	target-Sequence	0,10%	Generon - MODIfinder GMO SCREENING p35S /tNOS / pFMV Triplex			
15	06.07.			Gen-lal	FFS-Kit Promega	Real Time PCR, 45 cycles	
16	02.07.18	TARGET	0,01%	GENERON/ METHOD MI 034/17	DNA Extraction KITION FORCE GENERON	REAL TIME PCR	
17			23.01.00	biotecon	QIACube Automation Mericon Food Kit	Real time PCR	
18	05.07.18	=	≤ 0,01 %	SureFood® GMO SCREEN 4plex 35S/NOS/FMV+IAC (S2126), R-Biopharm / Congen	extraction by SureFood® PREP Basic Art. No. S1052	real-time PCR	-
19	05.07.18			foodproof GMO Screening Kit (35 S, NOS, bar, FMV) -5'Nuclease- from BIOTECON, Version 3 March 2017	foodproof GMO Sample Preparation Kit III from BIOTECON, Version 1 Sept. 2014	Real Time PCR	
20	05.06.18		0,05%	JRC/UERL/ISO/DIS 21570	CTAB method-200ng/µl	Real time PCR/45 cycles/82pb/ERM reference material	
21	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
22	18.06.18		20 copies	Swiss Book of Food Stuffs	Wizard DNA-extraction / 100 ng pro Sample	Real Time PCR / 50 Cycles	
23	06.06.18		0,01/	GEN-IAL Triplex I	1g, Simplex-Easy Spin Food kit /GEN-IAL	real-time PCR	
24	21.06.18	35 S	< 5 copies	R-Biopharm	R-Biopharm, SureFood PREP Advanced, as per kit instructions	Real Time PCR, SureFood GMO 4plex, R- Biopharm, as per kit instructions	
25	19/06	35S	ct-value 45	GEN-IAL genControl RT- Triplex IV p35S / NOS / pFMV, incl. IC	Congen SureFood PREP Basic extractionskit	Real Time PCR, 45 Cycles, Reference material ERM-BF 410dn	* traces (< 0,1%)
26	11.06.18	DNA	0.1%	inhouse	Wizard extraction	real time PCR	
27	22.06.	P-35-S		Huber et al. 2013	in house (CTAB and clean-up by innuPrep Micro Kit)	Real Time PCR/50 Cycles	

5.1.2 NOS-Screening Sequence

Evaluation number	Date of Analysis	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
NOS	Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA amount	e.g real time PCR / gel electrophoresis / cycles / amplificate length / reference material	
1	13.07.18		0,10%	In house method	Magnetic Bead Extraction	Gel Electrophoresis	
2	20.06.2018.	NOS	0.01	Biotecon Screening 1 Lyokit	Biotecon Sample Prep. Kit 3	Real-time PCR	
3	05.06.18					Real-time PCR	
4	13.06.18	180 bp		as per ASU § 64 L00.00-118 (primer sequence, PCR premix, PCR program)	DNA-extraction by Wizard-Kit from Promega	gel electrophoresis	
5	26.06.18	tNOS A. tumefaciens	LOD: 5 DNA-copies	Congen, Surefood GMO screen 35S, NOS & FMV (S2026)	Kit: Machery Nagel, Nucleospin DNA yield: >50 ng/µL A260/280: >1,6	qPCR, 45 cycles, reference material 1% CRM	Positive: >or = 0,9% Negative: <0,9%
6	11.07.18	nos	0,10%	Congen SureFood GMO 1 Kit	SureFood Prep Advanced Kit	Real-Time PCR	
7	11.07.18	target-Sequence	<0,01%	in House Method	prepman ultra applyed biosystem-no clean up- taqman universal master mix applyed biosystem	real time PCR	
8	28.06.18			GEN-IAL		Real Time PCR	
9	18.06.18			Biotecon		RealTime PCR	
10a	08.06.		10 copies	GEN-IAL GmbH	Genomic DNA from Food (Macherey-Nagel)	Real Time PCR	
10b	13.06.	DNA	0,03 %	ASU §64 method 00.00 122	in house method	Real Time PCR	
11	12.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
12							
13		Target / DNA	0,1%	Manufacturer Bioside / ISO 21569:2005/Amd 1:2013	Nucleo spin Food Macherey-Nagel	Real Time PCR	
14	06.07.18	target-Sequence	0,10%	Generon - MODIfinder GMO SCREENING p35S/tNOS/pFMV Triplex			
15	06.07.			Gen-lal	FFS-Kit Promega	Real Time PCR, 45 Zyklen	
16	02.07.18	TARGET	0,01%	GENERON/ METHOD MI 034/17	DNA Extraction KITION FORCE GENERON	REAL TIME PCR	
17			23.01.00	biotecon	QIACube Automation Mericon Food Kit	Real time PCR	
18	05.07.18	-	≤ 0,01 %	SureFood® GMO SCREEN 4plex 35S/NOS/FMV+IAC (S2126), R-Biopharm / Con- gen	extraction by SureFood® PREP Basic Art. No. S1052	real-time PCR	-
19	05.07.18						
20							
21	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
22	18.06.18		20 copies	Schweiz. Lebensmittelbuch	Wizard DNA-Extraktion / 100 ng pro Sample	Real Time PCR / 50 Cyclen	
23	06.06.18		0,01/	GEN-IAL Triplex I			
24	21.06.18	NOS	< 5 copies	R-Biopharm	R-Biopharm, SureFood PREP Advanced, as per kit instructions	Real Time PCR, SureFood GMO 4plex, R-Bio- pharm, as per kit instructions	
25	19/06	nos	ct-Wert 45	GEN-IAL genControl RT-Tri- plex IV p35S / NOS / pFMV, incl. IC	Congen SureFood PREP Basic extractionskit	Real Time PCR, 45 Cycles, Reference material ERM-BF 410dn	
26	11.06.18	DNA	0.1%	inhouse	Wizard extraction	real time PCR	
27	22.06.	T-nos		Huber et al. 2013	in house (CTAB and clean-up by innuPrep Micro Kit)	Real Time PCR/50 Cycles	

5.1.3 FMV-Screening Sequence

Evaluation number	Date of Analysis	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
FMV	Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA amount	e.g real time PCR / gel electrophoresis / cycles / amplificate length / reference material	
1							
2	20.06.2018.	FMV	0.01	Biotecon Screening 1 Lyokit	Biotecon Sample Prep. Kit 3	Real-time PCR	
3	05.06.18					Real-time PCR	
5	26.06.18	p34S FMV	LOD: 5 DNA-copies	Congen, Surefood GMO screen 35S, NOS & FMV (S2026)	Kit: Machery Nagel, Nucleospin DNA yield: >50 ng/µL A260/280: >1,6	qPCR, 45 cycles, reference material 1% CRM	Positive: >or = 0,9% Negative: <0,9% *) Amplification for FMV was detected but less then 0,9%
6	11.07.18	FMV	0,10%	Congen SureFood GMO 1 Kit	SureFood Prep Advanced Kit	Real-Time PCR	
7							
8	28.06.18			GEN-IAL		Real Time PCR	
9	18.06.18			Biotecon		RealTime PCR	
10							
10	13.06.	DNA	0,003 %	ASU §64 Method 00.00 148		Real Time PCR	
11	12.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
12							
13		Target / DNA	0,1%	Manufacturer Bioside / ISO/TS 21569-5:2016	Nucleo spin Food Macherey-Nagel	Real Time PCR	
14	06.07.18	target-Sequence	0,10%	Generon - MODIfinder GMO SCREENING p35S/tNOS/pFMV Triplex			
15	06.07.			Gen-lal	FFS-Kit Promega	Real Time PCR, 45 cycles	
16	28.06.18	TARGET	0,01%	GENERON/ METHOD MI 034/17	DNA Extraction KITION FORCE GENERON	REAL TIME PCR	
17			23.01.00	biotecon	QIACube Automation Mericon Food Kit	Real time PCR	
18	05.07.18	-	≤ 0,01 %	SureFood® GMO SCREEN 4plex 35S/NOS/FMV+IAC (S2126), R-Biopharm / Congen	extraction by SureFood® PREP Basic Art. No. S1052	real-time PCR	-
19	05.07.18						
20							
21	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
22	20.06.18		20 copies	Eugster et al	Wizard DNA-Extraction / 100 ng pro Sample	Real Time PCR / 50 Cycles	P-FMV was detected! Not the virus.
23	06.06.18		0,01/	GEN-IAL Triplex VII			
24	21.06.18	FMV	< 5 copies	R-Biopharm	R-Biopharm, SureFood PREP Advanced, as per kit instructions	Real Time PCR, SureFood GMO 4plex, R- Biopharm, as per kit instructions	
25	19/06	FMV	ct-Wert 45	GEN-IAL genControl RT- Triplex IV p35S / NOS / pFMV, incl. IC	Congen SureFood PREP Basic extractionskit	Real Time PCR, 45 Cycles, Reference material ERM-BF 410dn	
26	11.06.18	DNA	0.1%	inhouse	Wizard extraction	real time PCR	
27	22.06.	P-FMV		Mano et al 2009.	in house (CTAB and clean-up by innuPrep Micro Kit)	Real Time PCR/50 Cycles	

5.1.4 p-NOS/nptII Screening Sequence

Evaluation	Date of	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
number pNOS / nptll	Analysis Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA	e.g real time PCR / gel electrophores is / cycles / amplificate	
	Daymonar	ranger dequented / Bret	namber of depice? 707 of value	Manaradiard / Critical Method	amount	length / reference material	
1							
2	44.00.40		+			B 111 BOD	
3	14.06.18					Real-time PCR	
<u>4</u> 5							
5							
6	11.07.18	NPTII	0,10%	congen SureFood GMO 2 Kit	SureFood Prep Advanced Kit	Real-Time PCR	
7							
8							
9							
10							
11	12.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
12							
13							
14	06.07.18	target-Sequence	0,10%	Generon - MODIfinder GMO SCREENING NPTII/PAT/EPSPS/pNOS Quadruplex			
15	06.07.			Gen-lal	FFS-Kit Promega	Real Time PCR, 45 cycles	
16					•		
17							
18							
19							
20					·		
21	11.06.18	•	0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
22					·		·
23	13.06.18		0,01/	GEN-IAL Pnos-nptII			
24				·			
25		·					
26							
27							

5.1.5 CTP2-CP4 EPSPS Screening Sequence

Evaluation number	Date of Analysis	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
CTP2-CP4 EPSPS	Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA amount	e.g real time PCR / gel electrophoresis / cycles / amplificate length / reference material	
1							
2							
3	05.06.18					Real-time PCR	
4							
5							
6	11.07.18	CTPS:CP4 EPSPS	0,10%	congen SureFood GMO 2 Kit	SureFood Prep Advanced Kit	Real-Time PCR	
7							
8							
9							
10	08.06.		10 copies	GEN-IAL GmbH	Genomic DNA from Food (Macherey-Nagel)	Real Time PCR	
11	12.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
12							
13							
14	06.07.18	target-Sequence	0,10%	Generon - MODIfinder GMO SCREENING NPTII/PAT/EPSPS/pNOS Quadruplex			
15	06.07.			Gen-lal	FFS-Kit Promega	Real Time PCR, 45 cycles	
16							
17							
18	05.07.18	-	≤ 0,01 %	SureFood® GMO SCREEN 4plex BAR/NPTII/PAT/CTP2:CP4 EPSPS (S2127), R-Bio- pharm / Congen	Extraction with SureFood® PREP Basic Art. No. S1052	real-time PCR	-
19							
20							
21	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
22	20.06.18		20 copies	in house Method	Wizard DNA-Extraktion / 100 ng pro Sample	Real Time PCR / 50 Cycles	
23	06.06.18		0,01/	GEN-IAL Triplex I			
24						DLA2018	
25							
26	11.06.18	DNA	0.1%	inhouse	Wizard Extraction	real time PCR	
27	22.06.	ctp2-cp4-epsps		Huber et al 2013	in house (CTAB and clean-up by innuPrep Micro Kit)	Real Time PCR/50 Cycles	

5.1.6 35S-Pat Screening Sequence

Evaluation number	Date of Analysis	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
35S-Pat	Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA amount	e.g real time PCR / gel electrophoresis / cycles / amplificate length / reference material	
1							
2							
3	14.06.18					Real-time PCR	
4							
5							
6	11.07.18	PAT	0,10%	congen SureFood GMO 2 Kit	SureFood Prep Advanced Kit	Real-Time PCR	
7							
8							
9							
10							
11	12.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
12							
13							
14	06.07.18	target-Sequence	0,10%	Generon - MODIfinder GMO SCREENING NPTII/PAT/EPSPS/pNOS Quadruplex			
15	06.07.			Gen-lal	FFS-Kit Promega	Real Time PCR, 45 cycles	
16						` ′	
17			29.01.00		QIACube Automation Mericon Food Kit	Real time PCR	
18							
19							
20							
21	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
22							
23	06.06.18		0,01/	GEN-IAL Triplex VII			
24							
25							·
26							
27							

5.1.7 Cry1Ab/Ac Screening Sequence

Evaluation number	Date of Analysis	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
Cry1Ab/Ac	Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA amount	e.g real time PCR / gel electrophoresis / cycles / amplificate length / reference material	
1							
2							
3	12.07.18					Real-time PCR	
4							
5							
6							
7							
8							
9							
10							
11	27.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
12							
13							
14							
15	06.07.			Gen-lal	FFS-Kit Promega	Real Time PCR, 45 cycles	
16							
17							
18							
19							
20	15.06.18		0,05%	JRC/UERL/ISO/DIS 21570	CTAB method-200ng/µl	gel electrophoresis/35 cycles/189pb/ERM reference material	
21	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
22							
23	13.06.18		0,01/	GEN-IAL Duplex cry1Ab/Ac- Pnos			
24							
25							
26		<u> </u>					·
27		·					·

5.1.8 GMO-Maize (bt11)

Evaluation number	Date of Analysis	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
GMO-Maize (bt11)	Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA amount	e.g real time PCR / gel electrophoresis / cycles / amplificate length / reference material	
1	13.07.18		0,10%	In house Method	Magnetic Bead Extraction	Gel Electrophoresis	
2							
3	05.06.18					Real-time PCR	
4	25.06.18			in house method	DNA-extraction by Wizard-Kit from Promega	gel electrophoresis	
5							
6							
7							
8							
9							
10							
11	27.06.18	Target-DNA	0.1 %	Eurofins	biotecon sample preparation III	Real Time PCR	
12							
13							
14							
15	06.07.			Gen-lal	FFS-Kit Promega	Real Time PCR, 45 cycles	
16							
17							
18	05.07.18	-	≤ 0,01 %	SureFood® GMO QUANT Bt11 Corn (S2016), R-Bio- pharm / Congen	Extraction with SureFood® PREP Basic Art. No. S1052	real-time PCR	-
19							
20							
21	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
22	21.06.18	·	20 copies	in house Method	Wizard DNA-Extraction / 100 ng per Sample	Real Time PCR / 50 Cycles	
23	07.06.18		0,01/	GEN-IAL GMO-com			
24							
25							
26	11.06.18	DNA	0.1%	inhouse	Wizard Extraction	real time PCR	
27	28.06.	IBR		JRC 2008 Event specific Me- thod QT-EVE-ZM-015	in house (CTAB and clean-up with innuPrep Micro Kit)	Real Time PCR/50 cycles	

5.1.9 GMO-Maize (MIR604)

Evaluation number	Date of Analysis	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
GMO-Maize (MIR604)	Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA amount	e.g real time PCR / gel electrophoresis / cycles / amplificate length / reference material	
1							
2							
3	11.06.18					Real-time PCR	
4							
5							
6							
7							
8							
9							
10							
11	27.06.18	Target-DNA	0.1 %	Eurofins	biotecon sample preparation III	Real Time PCR	
12							
13							
14							
15	06.07.			Gen-lal	FFS-Kit Promega	Real Time PCR, 45 cycles	
16							
17							
18	05.07.18	-	≤ 0,01 %	in house method	Extraction with SureFood® PREP Basic Art. No. S1052	real-time PCR	-
19							
20							
21	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
22							
23	11.06.18		0,01/	GEN-IAL RT-MIR604			
24							
25							
26	11.06.18	DNA	0.1%	inhouse	Wizard Extraction	real time PCR	
27	28.06.	IBR		JRC 2007 Event specific Me- thod QT-EVE-ZM-013	in house (CTAB and clean-up with innuPrep Micro Kit)	Real Time PCR/50 cycles	

5.1.10 Maize-DNA (Maize specific)

Evaluation number	Date of Analysis	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
Maize DNA	Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA amount	e.g real time PCR / gel electrophoresis / cycles / amplificate length / reference material	
1	13.07.18	Invertase	0,10%	In house method	Magnetic Bead Extraction	Gel Electrophoresis	
2							
3	13.07.18					Real-time PCR	
4	26.06.18	226 bp		as per ASU § 64 L00.00-118 (primer sequence, PCR premix, PCR program)	DNA-extraction by Wizard-Kit from Promega	gel electrophoresis	
5							
6							
7	11.07.18	target-Sequence	<0,01%	in House Method	prepman ultra applyed biosystem-no clean up- taqman universal master mix applyed biosystem	real time PCR	Zeina
8							
9							
10							
11	27.06.18	Target-DNA	0.1 %	Eurofins	biotecon sample preparation III	Real Time PCR	
12							
13							
14							
15	06.07.			Gen-lal	FFS-Kit Promega	Real Time PCR, 45 cycles	
16							
17							
18	05.07.18	-	≤ 4 mg/kg	SureFood® GMO Plant 4plex Com/Soya/Canola+IAC (S2158), R-Biopharm / Con- gen	extraction by SureFood® PREP Basic Art. No. S1052	real-time PCR	-
19							
20							
21	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
22	18.06.18		20 copies	in house Method	Wizard DNA-Extraction / 100 ng per Sample	Real Time PCR / 50 Cycles	
23							
24							
25							
26	11.06.18	DNA	0.1%	inhouse	Wizard Extraction	real time PCR	
27	28.06.	adh1		L.00.00-105	in house (CTAB and clean-up by innuPrep Micro Kit)	Real Time PCR/50 Cycles	

5.1.11 GMO-Soya RR (GTS 40-3-2)

Evaluation number	Date of Analysis	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
GM-Soya RR (GTS 40-3-2)	Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA amount	e.g real time PCR / gel electrophoresis / cycles / amplificate length / reference material	
1	13.07.18		0,10%	In house method	Magnetic Bead Extraction	Gel Electrophoresis	
2							
3	12.06.18					Real-time PCR	
4	15.06.18	172 bp		as per ASU § 64 L00.00-118 (primer sequence, PCR premix, PCR program)	DNA-extraction by Wizard-Kit from Promega	gel electrophoresis	
5							
6							
7							
8							
9							
10a	27.06.		8 copies	GEN-IAL GmbH	Genomic DNA from Food (Macherey-Nagel)	Real Time PCR	
10b	12.06.	DNA	0,05 %	ASU §64 Method 00.00 105	in house method	Real Time PCR	
11	14.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
12							
13							
14							
15	06.07.			Gen-lal	FFS-Kit Promega	Real Time PCR, 45 cycles	
16							
17							
18	05.07.18	-	≤ 0,01 %	SureFood® GMO ID Roun- dup Ready Soya (S2030), R- Biopharm / Congen	extraction by SureFood® PREP Basic Art. No. S1052	real-time PCR	-
19							
20	13.06.18		0,05%	JRC/UERL/ISO/DIS 21570	CTAB method-200ng/µl	gel electrophoresis/35 cycles/169pb/ERM reference material	
21	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
22	21.06.18		20 copies	Swiss Book of Food Stuffs	Wizard DNA-extraction / 100 ng pro Sample	Real Time PCR / 50 Cycles	
23	07.06.18		0,01/	GEN-IAL GMO-soy			
24		·					·
25							
26	11.06.18	DNA	0.1%	inhouse	Wizard Extraction	real time PCR	
27	28.06.	IBR		L.00.00-105	in house (CTAB and clean-up by innuPrep Micro Kit)	Real Time PCR/50 Cycles	

5.1.12 GMO-Soya RR2 (MON89788)

Evaluation number	Date of Analysis	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
GM Soya RR2 (MON89788)	Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA amount	e.g real time PCR / gel electrophoresis / cycles / amplificate length / reference material	
1							
2							
3	05.06.18					Real-time PCR	
4	09.07.18	139 bp		Nach Charles Delobel C. Et al. (2013): Event-specific Method for the Quantification of Soybean Line MON 89788 Using Real-time PCR v1.01 - Validation Report and Vali- dated Method	DNA-extraction by Wizard-Kit from Promega	gel electrophoresis	
5							
6							
7							
8							
9							
10	27.06.		2 copies	GEN-IAL GmbH	Genomic DNA from Food (Macherey-Nagel)	Real Time PCR	
10	12.06.	DNA	0,0015 %	JRC (QT-EVE-GM-006)	in house method	Real Time PCR	
11	13.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
12							
13							
14							
15	06.07.			Gen-lal	FFS-Kit Promega	Real Time PCR, 45 cycles	
16							
17							
18	05.07.18	-	≤ 0,01 %	SureFood® GMO ID RR2Y Soya (S2034), R-Biopharm / Congen	extraction by SureFood® PREP Basic Art. No. S1052	real-time PCR	-
19							
20	· ·						
21	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
22	21.06.18		20 copies	EU-RL GMFF-Method	Wizard DNA-extraction / 100 ng pro Sample	Real Time PCR / 50 Cycles	
23	07.06.18		0,01/	GEN-IAL GMO-soy			
24							
25							
26	11.06.18	DNA	0.1%	inhouse	Wizard extraction	real time PCR	
27	28.06.	IBR		JRC 2013 Event specific Me- thod QT-EVE-GM-006	in house (CTAB and clean-up by innuPrep Micro Kit)	Real Time PCR/50 Cycles	

5.1.13 Lectin-DNA

Evaluation number	Date of Analysis	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
Lectin DNA	Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA amount	e.g real time PCR / gel electrophoresis / cycles / amplificate length / reference material	
1	13.07.18		0,10%	In house Method	Magnetic Bead extraction	Gel electrophoresis	
2							
3	13.07.18					Real-time PCR	
4	15.06.18	118 bp		as per ASU § 64 L00.00-118 (primer sequence, PCR premix, PCR program)	DNA-extraction by Wizard-Kit from Promega	gel electrophoresis	
5							
6							
7							
8							
9							
10							
11	14.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
12							
13							
14							
15	06.07.			Gen-lal	FFS-Kit Promega	Real Time PCR, 45 cycles	
16							
17							
18	05.07.18	-	≤ 5 DNA-copies	SureFood® GMO QUANT Roundup Ready Soya (S2014), R-Biopharm / Con- gen	extraction by SureFood® PREP Basic Art. No. S1052	real-time PCR	-
19							
20	05.06.18		0,10%	JRC/UERL/ISO/DIS 21570	CTAB method-200ng/µl	Real time PCR/45 cycles/81pb/ERM reference material	
21	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
22	18.06.18		20 copies	Swiss Book of Food Stuffs	Wizard DNA-extraction / 100 ng pro Sample	Real Time PCR / 50 Cycles	
23							
24							
25							
26	11.06.18	DNA	0.1%	inhouse	Wizard extraction	real time PCR	
27a	28.06.	lectin (Le1) gene		ISO 21570:1-103 (2005); JRC Method QT-TAX-GM- 002;	in house (CTAB and clean-up by innuPrep Micro Kit)	Real Time PCR/50 Cycles	
27b	28.06.	IBR		L.00.00-105	in house (CTAB and clean-up by innuPrep Micro Kit)	Real Time PCR/50 Cycles	

5.1.14 Other DNA-Sequences

Parameter	Evaluation number	Date of Analysis	Results given as	Limit of Detection	Test-Kit or Literature	Notes to Extraction	Notes to PCR-reaction	Further Remarks
		Day/Month	Target-Sequence / -DNA	number of copies / % / ct-value	Manufacturer / Official Method	e.g. Extraction / enzymes / clean-up / DNA quality / DNA amount	e.g real time PCR / gel electrophoresis / cycles / amplificate length / reference material	
NK603	3a	12.06.18					Real-time PCR	NK603
CaMV	3b						Real-time PCR	CaMV
BAR	6	11.07.18	BAR	0,10%	congen SureFood GMO 2 Kit	SureFood Prep Advanced Kit	Real-Time PCR	BAR
Plant	10a	12.06.	DNA	0,02 %	in house method (Screening Plant-DNA)	in house method	Real Time PCR	
pat	10b	27.06.		8 copies	GEN-IAL GmbH	Genomic DNA from Food (Macherey-Nagel)	Real Time PCR	
bar	10c	27.06.		4 copies	GEN-IAL GmbH	Genomic DNA from Food (Macherey-Nagel)	Real Time PCR	
bar	11a	12.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
p-35-nptII / MON89034	11b	12.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
TC1507 Maize	11c	27.06.18	Target-DNA	0.1 %	Eurofins	biotecon sample preparation III	Real Time PCR	
A2704-12 Soya	11d	27.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
NK603 / MON810	11e	27.06.18	Target-DNA	0.1 %	biotecon	biotecon sample preparation III	Real Time PCR	
bar	19	05.07.18						
CaMV	21a	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
GVO-Maize (NK603)	21b	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
GVO-Maize (MON88017)	21c	11.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
SoyaA5547	21d	26.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
SoyaA2704	21e	26.06.18		0,01 %	in house method	CTAB-NuceloSpin	Real Time PCR; 45 cycles	
T-35S	22a	19.06.18		20 copies	Eugster et al	Wizard DNA-Extraction / 100 ng per Sample	Real Time PCR / 50 cycles	
bar	22b	20.06.18		20 copies	in house method	Wizard DNA-Extraktion / 100 ng pro Sample	Real Time PCR / 50 cycles	
PAT	22c	20.06.18		20 copies	in house method	Wizard DNA-Extraktion / 100 ng pro Sample	Real Time PCR / 50 cycles	
GOX	22d	20.06.18		20 copies	in house method	Wizard DNA-Extraction / 100 ng per Sample	Real Time PCR / 50 cycles	
T25	23a			0,01/				to verify the positive pat detection further PCRs were applied: TC1507, DAS59122, T25. For T25 sample 1 was positive.
bar	23	06.06.18		0,01/	GEN-IAL Triplex VII			·
MON88017- Maize	23c	12.06.18		0,01/	GEN-IAL GMO-com			
NK603- Maize	23d	12.06.18		0,01/	GEN-IAL RT-NK603			
A2704-Soya	23e	12.06.18		0,01/	GEN-IAL Triplex-Soya I			
A5547-Soya		12.06.18		0,01/	GEN-IAL Triplex-Soya I			
Pat	26a	11.06.18	DNA	0.1%	in house method	Wizard Extraction	real time PCR	Pat
NK603	26b		DNA	0.1%	in house method	Wizard Extraction	real time PCR	NK603
TC1507	26c		DNA	0.1%	in house method	Wizard Extraction	real time PCR	TC1507

5.2 Homogeneity

5.2.1 Mixture homogeneity before bottling

Microtracer Homogeneity Test

DLA 34-2018 Sample 1

Result of analysis

Sample	Weight [g]	Particle number	Particles [mg/kg]
1	4,98	49	19,7
2	4,98	48	19,3
3	5,05	47	18,6
4	4,99	52	20,8
5	5,02	51	20,3
6	4,99	51	20,4
7	4,99	52	20,8
8	5,06	50	19,8

8 7	
7	
50,0	Particles
1,96	Particles
0,54	
100	%
112	%
	0,54 100

Normal distribution		
Number of samples	8	
Mean	20,0	mg/kg
Standard deviation	0,78	mg/kg
rel. Standard deviaton	3,93	%
Horwitz standard deviation	10,2	%
HorRat-value	0,39	
Recovery rate	112	%

Microtracer Homogeneity Test DLA 34-2018 Sample 2

Result of analysis

Sample	Weight [g]	Particle number	Particles [mg/kg]
1	5,07	42	16,6
2	5,03	45	17,9
3	4,97	58	23,3
4	5,05	56	22,2
5	5,01	61	24,4
6	4,98	42	16,9
7	4,97	60	24,1
8	5,05	46	18,2

Poisson distribution		
Number of samples	8	
Degree of freedom	7	
Mean	51,3	Particles
Standard deviation	8,46	Particles
χ² (CHI-Quadrat)	9,77	
Probability	20	%
Recovery rate	97	%

Normal distribution		
Number of samples	8	
Mean	20,4	mg/kg
Standard deviation	3,37	mg/kg
rel. Standard deviaton	16,5	%
Horwitz standard deviation	10,2	%
HorRat-value	1,6	
Recovery rate	97	%

Microtracer Homogeneity Test DLA 34-2018 Sample 3

Result of analysis

Sample	Weight [g]	Particle number	Particles [mg/kg]
1	5,02	52	20,7
2	4,96	49	19,8
3	5,02	51	20,3
4	5,01	44	17,6
5	4,97	44	17,7
6	5,03	48	19,1
7	5,03	51	20,3
8	5,03	49	19,5

8	
7	
48,5	Particles
2,96	Particles
1,27	
99	%
81	%
	7 48,5 2,96 1,27 99

Normal distribution		
Number of samples	8	
Mean	19,4	mg/kg
Standard deviation	1,18	mg/kg
rel. Standard deviaton	6,11	%
Horwitz standard deviation	10,2	%
HorRat-value	0,60	
Recovery rate	81	%

Microtracer Homogeneity Test DLA 34-2018 Sample 5

Result of analysis

Sample	Weight [g]	Particle number	Particles [mg/kg]
1	5,03	52	20,7
2	4,99	53	21,2
3	5,02	51	20,3
4	5,05	59	23,4
5	4,98	70	28,1
6	4,99	63	25,3
7	4,98	57	22,9
8	5,07	52	20,5

Particles
Particles
%
%

Normal distribution		
Number of samples	8	
Mean	22,8	mg/kg
Standard deviation	2,75	mg/kg
rel. Standard deviaton	12,1	%
Horwitz standard deviation	10,0	%
HorRat-value	1,2	
Recovery rate	112	%

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 34-2018		
PT name	GMO-Screening qualitative: 5 Samples with positive/negative amounts of Screening Targets 35S, NOS, FMV, CTP2-CP4 EPSPS / GMO-Maize (Bt11, MIR604) and GMO-Soya (RR GTS 40-3-2, RR2 MON89788)		
Sample matrix*	Five different Samples: possible ingredients: Products of soybean, maize and wheat flour and semolina		
Number of samples and sample amount	Five different samples, 10 g each.		
Storage	Samples: dry and dark at room temperature (long term cooled 2 - 10°C)		
Intentional use	Laboratory use only (quality control samples)		
Parameter	qualitative: Target sequences 35S, NOS, FMV, CTP2-CP4 EPSPS as well as GMO-maize (Bt11, MIR604) and GMO-soya (RR GTS 40-3-2, RR2 MON89788)		
Methods of analysis	Analytical methods are optional		
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 1-5 per parameter and filled in the result submission file		
Units	positive / negative (limit of detection: copies or percentage)		
Number of significant digits	only qualitative		
Further information	Further information can be given in the result submission file.		
Result submission	The result submission file should be sent by e-mail to: pt@dla-lvu.de		
Deadline	the latest 13 th July 2018		
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.		

^{*} Control of mixture homogeneity and qualitative testings are carried out by DLA. 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
		AUSTRIA
		Germany
		AUSTRIA
		Germany
		Germany
		Germany
		SWITZERLAND
		SWITZERLAND
		Germany
		ITALY
		SPAIN
		ITALY
		ITALY
		BELGIUM
		Germany
		GREAT BRITAIN
		Germany
		Germany
		ITALY
		GREAT BRITAIN
		CROATIA
		Germany

[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

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