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Comparative Testing Report on the Detection and Quantification of GM Events in Rice Noodles

Comparative testing round: ILC-EURL-GMFF-CT-02/13

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Abstract

The European Union Reference Laboratory for Genetically Modified Food and Feed (EU-RL GMFF), accredited under ISO/IEC 17043, organised a comparative testing (CT) round for National Reference Laboratories (NRLs) nominated under Regulation (EC) No 882/2004, with voluntary participation of other official control laboratories.

The test items consisted of rice noodles and commercial soybeans spiked with ground powder of soybean event DP-356043-5 in two different concentrations (Level 1 and 2). Participants were required to perform species identification and test for the presence of any GM event in the two test items. Any event detected then had to be quantified. Participants could report the results in mass/mass % or copy/copy % and the EU-RL GMFF calculated the robust means (ØR) for Level 1 and 2 test items accordingly. The target standard deviation for CT was fixed by the Advisory Board for Comparative Testing at 0.2 for the event, based on the experience of previous CT rounds. The robust means and target standard deviation were used to derive z-scores for the participants' results.

Eighty-eight laboratories from 42 countries registered for this CT round, of which 85 laboratories from 41 countries returned at least qualitative test results.

When performing species identification, almost all laboratories correctly identified soybean and rice in both test items, and a few laboratories also detected maize and/or oilseed rape. In total 71 laboratories reported the presence of GM material in the test items, but 14 laboratories failed in this task. All of the 71 laboratories, except eight, correctly identified soybean event DP-356043-5 in the test items.

Results of the quantitative evaluation of the GM content were satisfactory for both measurement units, with only two NRLs appointed under Regulation (EC) No 1981/2006 (one measuring in m/m % and one in cp/cp %) obtaining unsatisfactory z-scores ($|z| \ge 2.0$) for both test items.

Despite the overall satisfactory outcome of this CT round, only 58 % of participants provided information on measurement uncertainty in a complete and consistent manner, and further improvement in this crucial area is needed.





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Confidentiality statement: The laboratory codes assigned to each participant in this comparative testing round are confidential. However, the EU-RL GMFF will disclose details of the National Reference Laboratories that have been appointed under Regulation (EC) No 882/2004 to DG SANCO.

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EU-RL GMFF: Comparative testing report

Executive Summary

The European Union Reference Laboratory for Genetically Modified Food and Feed (EU-RL GMFF), accredited under ISO/IEC 17043, organised a comparative testing (CT) round for National Reference Laboratories (NRLs) nominated under Regulation (EC) No 882/2004, with voluntary participation of other official control laboratories.

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1. Introduction

The Joint Research Centre (JRC) of the European Commission was established as European Union Reference Laboratory for GM Food and Feed by Regulation (EC) No 1829/2003⁽¹⁾. The EU-RL GMFF is also mandated by Regulation (EC) No 882/2004⁽²⁾.

Article 32 of Regulation (EC) No 882/2004 tasks the EU-RLs with the organisation of comparative testing (CT) for National Reference Laboratories (NRLs, nominated under Regulation 882/2004) and an appropriate follow-up of such testing. The aim of this activity is 'to contribute to a high quality and uniformity of analytical results'⁽²⁾. Moreover, Article 12 of the said Regulation requires that the nominated NRLs should be accredited under ISO/IEC 17025 on 'General requirements for the competence of testing and calibration laboratories' and 17025-accredited laboratories must prove their competence, *e.g.* by taking part in comparative testing. The EU-RL GMFF is accredited under ISO/IEC 17043⁽³⁾ to organise CT rounds.

Regulations (EC) No 1829/2003 and (EU) No 619/2011 establish a threshold for labelling of food and feed products (0.9 %) and a minimum required performance limit (0.1 % m/m) for detecting low level presence of GMO in feed. These values are used by the Member States of the European Union in the official control of food and feed. Hence, an accurate determination of the GM content is of paramount importance.

The EU-RL GMFF organised a comparative testing round for NRLs nominated under Regulation (EC) No 882/2004. Participation was open and free of charge for any official control laboratory. Participation was mandatory for NRLs nominated under Regulation (EC) No 882/2004 and highly recommended for NRLs nominated under Regulation (EC) No 1981/2006⁽³⁾. This comparative testing round met the requirements of ISO/IEC 17043.

In March 2014, a total of 155 laboratories were invited to participate in this CT round of the EU-RL GMFF (ILC-EURL-GMFF-CT-02/13) and 88 laboratories from 42 countries registered for it. Test items were prepared by the EU-RL and shipped to registered participants in mid-April 2014 in plastic containers containing approximately 10 g of flour. The EU-RL GMFF managed the on-line registration and submission of results and was responsible for their evaluation. It was supported by the Advisory Board for CT.

Eighty-five laboratories from 41 countries returned at least qualitative results (see Figures 1 and 2). These laboratories fell into the following groups:

- 1. 2 were NRLs nominated only under Regulation (EC) No 882/2004 (group 1),
- 2. 23 were NRLs nominated only under Regulation (EC) No 1981/2006 (group 2),
- 3. 28 were NRLs nominated under both Regulations (group 3),
- 4. 5 were ENGL members but did not belong to group 1, 2 or 3 (group 4),
- 5. 9 were official control laboratories from EU Member States but not ENGL members (group 5),
- 6. 18 were official control laboratories from a third country (group 6).

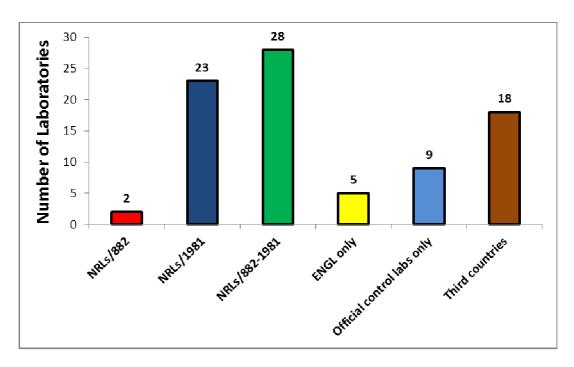


Figure 1. Laboratories submitting at least qualitative results, divided by group.

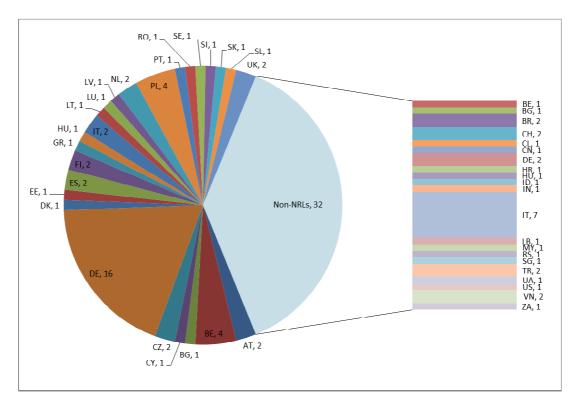


Figure 2. Overview of laboratories submitting at least qualitative results, divided by country.

2. Test items

The test items were produced in-house by the EU-RL GMFF from ground powder of DP-356043-5, provided by IRMM, Geel (Belgium) and rice noodles and soybeans purchased at the local market.

2.1 Characterisation of base materials

Base materials consisted of:

- 6000 g of non-GM rice noodles
- 500 g of non-GM soybeans
- 300 g of GM soybean event DP-356043-5 powder provided by IRMM, Geel, Belgium

Non-GM materials were ground using an Ultra Centrifugal Mill ZM200 (Retsch GmbH, DE). An oven-drying method was used for determining the remaining water content in the powders. To determine the extractability of DNA from the GM and non-GM base materials, DNA was extracted from each of the powders in 10 independent replicates using a modified CTAB method. Extracted DNA was quantified with Picogreen in a VersaFluor Fluorometer.

Four DNA extracts, randomly chosen from the 10 replicates, were assessed for the presence of inhibitors using the validated *le1* reference gene system and the amount of DNA validated for the event-specific method for DP-356043-5 soybean. No inhibition was detected. The DNA extracts were also assessed for the presence of GM-event(s) or species-specific DNA other than those relevant to the present comparative testing round, using ABI pre-spotted plates⁽⁵⁾. No other species were identified in the base materials.

2.2 Preparation and characterisation of test items

Two levels of processed material (Level 1 and 2) test items were gravimetrically prepared to obtain nominal concentration values of 0.7 m/m % and 1.5 m/m % DP-356043-5 soybean.

These test items were prepared by the EU-RL GMFF in accordance with ISO Guide 34⁽⁶⁾ ('General requirements for the competence of reference material producers'), as follows:

- Two different mass fractions (mixtures) of the GM material, representing two different GM levels, were produced by mixing pure non-GM with pure GM powder base materials, taking into account the water content of the base materials (see Table 1 for details on mixtures);
- Each mixture was manually mixed for 10 minutes, then thoroughly mixed for 60 min in a Turbula T10B mixer.

Table 1. Mixtures composition, in g.

Took its was	No	n-GM	GM
Test items	Noodles flour	Non-GM soybean	DP-356043-5 soybean
Level 1	2307.12	49.22	0.31
Level 2	2307.12	48.83	0.67

From each of these two powder test materials, 200 test items of up to 10 g were prepared in 30 ml-bottles using a sample divider (Retsch GmbH, Haan, DE). Bottles were labelled according to the GM level of the test items and stored at 4° C.

Homogeneity and stability testing of test items was performed in-house. Both test items were found to be homogeneous for the GM event (p-value > 0.05), and they were found to remain stable over a time period of 4 weeks at a 5 % significance level. Details of the testing performed are described in Annex 1.

3. Tasks to be performed by participants

Participants in this CT round were required to analyse the two test items (Level 1 and 2), i.e.:

- Perform species identification: maize, soybean, oilseed rape and rice;
- Identify and quantify the GM event(s) detected.

Participants could report the quantitative results in m/m % or DNA cp/cp %. Additionally, laboratories were asked to report the estimated measurement uncertainty as an absolute value, and the practical LOD and LOQ in the appropriate measurement unit.

Participants were instructed to apply the formulas described below when reporting their results:

Target taxon-specific DNA copy numbers [cp]

4. Results

A total of 85 laboratories from 41 countries submitted at least answers to the questionnaire and results from species identification. Of these, 71 reported the presence of GM material at least for one test item and 63 correctly identified soybean event DP-356043-5 in at least one test item. Quantitative results for this event were submitted by 56 and 57 laboratories for test items 1 and 2, respectively.

4.1 Species identification

Overall, the majority of the 85 laboratories performing at least species identification correctly detected the presence of rice and soybean (80 and 85 for Level 1, 78 and 81 for Level 2; see Figures 3a and 3b) and the absence of maize and oilseed rape (76 and 73 for Level 1, 72 and 69 for Level 2). Few laboratories detected maize in the test items (8 and 11 for Level 1 and 2, respectively). Additionally, the following laboratories did not test for rice or oilseed rape (5 and 11 laboratories, respectively):

- No testing for rice: one NRL appointed under both Regulations 882/2004 and 1981/2006 and 4 non-NRLs;
- No testing for oilseed rape: one NRL appointed under Regulation 1981/2006 only and 10 non-NRLs.

Some discordant results between test item Level 1 and 2 were reported, possibly because the results were inadvertently entered incorrectly for the two test items; this may be caused by the fact that the order of the selection options was different between the questions (first and second options to select were "Presence" and "Absence" for Level 1 and "Absence" and "Presence" for Level 2). Four laboratories provided different results for both test items, one NRL appointed under Regulations 882/2004 and 1981/2006, one NRL appointed under Regulation 1981/2006 only and two official control laboratories from outside the EU. Results for all species, regardless of the laboratory category, are summarised in Figures 3a-b, and detailed results are reported in Annex 2.

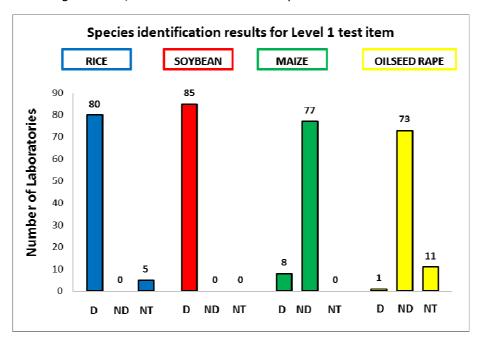


Figure 3a. Overview of species identification data for Level 1 test item. D = Detected, ND = Not Detected, NT = Not Tested.

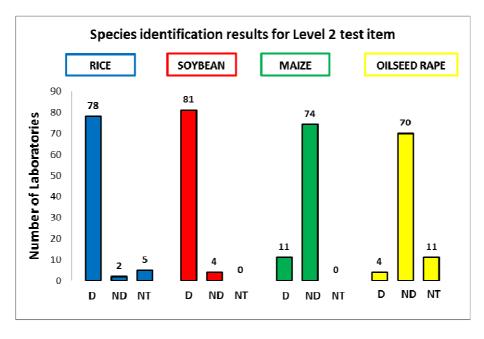


Figure 3b. Overview of species identification data for Level 2 test item. D = Detected, ND = Not Detected, NT = Not Tested.

4.2 GM detection

Of the 85 laboratories performing species identification, 71 reported information on the detection of GM material, *i.e.* they detected the presence of either p35S or T-*nos* or both GM elements. The remaining 14 laboratories, listed in Table 2, did not detect any GM element or GM event. Among these 14 laboratories, there were 3 NRLs, 5 official control laboratories from within the EU and 6 from third countries.

Table 2. Laboratories that performed species identification but did not report any GM event or GM element.

Laboratory number	Group	
L09	3	(NRLs/882-1981)
L13	3	(NRLs/882-1981)
L40	2	(NRLs/1981)
L06	5	(Off contr EU)
L14	5	(Off contr EU)
L79	5	(Off contr EU)
L80	5	(Off contr EU)
L84	5	(Off contr EU)
L12	6	(Off contr non-EU)
L39	6	(Off contr non-EU)
L41	6	(Off contr non-EU)
L49	6	(Off contr non-EU)
L51	6	(Off contr non-EU)
L54	6	(Off contr non-EU)

4.3 GM event(s) identification

Of the 71 laboratories that detected GM material, 62 and 63 correctly identified soybean event DP-356043-5 in Level 1 and 2 test items, respectively. Of the laboratories identifying events other than DP-356043-5, one NRL appointed under Regulation 882/2004 only identified MON 810 maize and two laboratories, including one NRL appointed under Regulation 1981/2006, only identified 40-3-2 (RR) soybean. Five other official control laboratories, two from the EU and three from third countries, only reported the presence of GM elements (p35S, T-nos) without performing further GM event identification (Table 3).

Table 3. Laboratories identifying GM events other than soybean event DP-356043-5 or only reporting GM elements (names of events and elements are as reported by the participants).

Laboratory	Group		Event/	Event/
number			Element 1	Element 2
L35	1	(NRLs/882)	MON810	-
L32	2	(NRLs/1981)	RR soy	-
L36	5	(Off contr EU)	35S	-
L66	5	(Off contr EU)	P35S	NOS
L63	6	(Off contr non-EU)	P35S	-
L69	6	(Off contr non-EU)	P35S	-
L82	6	(Off contr non-EU)	P35S	-
L83	6	(Off contr non-EU)	Soybean Line GTS 40-3-2	-

4.4 GM event(s) quantification

4.4.1 Quantitative results from the participants

Among the laboratories that correctly identified soybean event DP-356043-5, only 56 and 57 provided quantitative results for Level 1 and 2 test items, respectively. Most laboratories (approximately 85 % and 84 % for Level 1 and 2, respectively) reported the GM content of test items in m/m %, whereas the remaining laboratories expressed their results in cp/cp % (see Figure 4). Additionally, three laboratories submitting results in cp/cp % provided only qualitative results, *i.e.* presence below/above a certain threshold value, two of them for both test items and one only for Level 1.

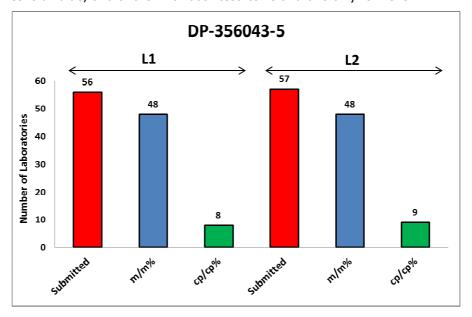


Figure 4. Overview of quantitative results for soybean event DP-356043-5, grouped by measurement unit. L1 = Level 1, L2 = Level 2.

One laboratory (L35) that did not identify soybean event DP-356043-5 (NRL appointed under Regulation 882/2004 only) provided quantitative results for maize event MON 810 for both test items (18.78 and 53.98 cp/cp %, respectively).

4.4.2 Consensus value from participants

The consensus value (μ_R) for DP-356043-5 from participants in the CT round was calculated using robust statistics^(7,8). This approach minimises the influence of outlying values. Robust means (μ_R) were calculated separately for measurement results reported in m/m % and cp/cp %.

The expanded uncertainty (U) comprises standard uncertainty (u) contributions from the characterisation of the material (u_{char}) and the between-test item homogeneity (u_{bb})⁽⁹⁾, and is estimated according to:

$$U = k\sqrt{u_{char}^2 + u_{bb}^2}$$
 (3)

A coverage factor (k) of 2 was used to calculate the expanded uncertainty corresponding to a 95 % level of confidence⁽¹⁰⁾. The standard uncertainty (u_{char}) on the characterisation was calculated using the formula:

$$u_{char} = \frac{\sigma}{\sqrt{N}}$$
 (4)

where: σ = robust Relative Standard Deviation of the robust mean expressed in m/m %

N = number of data points

The robust means (μ_R) for data on the non-transformed scale, and associated uncertainties, as calculated by the EU-RL GMFF, are reported in Table 4.

Table 4. Overview of robust means (μ_R) and expanded uncertainties for Level 1 and 2 test items.

Test item	μ _R [m/m %]	Expanded uncertainty (U = 2 * u)		Relative standard uncertainty contribution [%]			
		U abs [m/m %]	U rel [%]	(u _{char, rel}) ¹	(u _{bb, rel}) ²		
		DP-35	6043-5				
Level 1	0.57 (N = 48)	0.06	10.74	4.56	2.84		
Level 2	1.35 (N = 48)	0.20	14.47	4.34	5.79		
Test item	μ _κ [cp/cp %]	•	uncertainty 2 * u)	Relative standa contribut	,		
		U abs [cp/cp %]	U rel [%]	(u _{char, rel}) ¹	(u _{bb, rel}) ²		
	DP-356043-5						
Level 1	0.64 (N = 8)	0.16	25.21	12.28	2.84		
Level 2	1.37 (N = 9)	0.22	16.36	5.78	5.79		

¹ Relative standard uncertainty relating to the characterisation

4.4.3 Laboratories' performance

The z-scores were calculated for both Level 1 and 2 test items on the basis of the robust means for both m/m % and cp/cp % data (see Annex 3, formula A3.1). For consistency, all decimal numbers were rounded to two digits. Detailed results are reported in Annex 4, Tables A4.1 to A4.4 and Figures A4.1 to A4.4. Laboratories with a z-score outside the acceptable range (i.e. $|z| \ge 2.0$) are highlighted in bold in the tables.

In these tables, "Value" refers to the reported value and "Uncertainty" as calculated and reported by the laboratory. Also practical "LOD" (limit of detection) and practical "LOQ" (limit of quantification) are values calculated and provided by the laboratories and refer to the methods they used on the specific samples. In addition to the z-scores, the percentage of laboratories with incorrectly reported measurement uncertainty (MU; detailed in Section 5.2), the mean LOD (μ_{LOD}), mean LOQ (μ_{LOQ}) as well as their standard deviations and the fraction of laboratories outside the acceptable range of the z-score were calculated by the EU-RL GMFF and reported at the bottom of each table in Annex 4.

To facilitate the comparison between the groups of laboratories defined on page 6, the results reported in Annex 4 are stratified according to the following three categories:

- Category (a): NRLs appointed only under Regulation (EC) No 882/2004 and those appointed under both Regulations (groups 1 and 3),
- Category (b): NRLs appointed only under Regulation (EC) No 1981/2006 (group 2),
- Category (c): ENGL members (not in group 1, 2, or 3), non-ENGL EU laboratories and third countries official control laboratories (groups 4, 5, and 6).

² Relative standard uncertainty resulting from the homogeneity assessment

In Table 5 a summary of the results with respect to the indicators described above (LOD, LOQ, % of incorrectly reported MU and % of unsatisfactory z-scores) is presented, separately for the two measurement units and pooled across laboratory category.

Approximately one third of all laboratories incorrectly reported the measurement uncertainties. Among all laboratories submitting quantitative results for soybean event DP-356043-5, two laboratories (4%), both NRLs under Regulation 1981/2006 only, obtained z-scores outside the acceptable range for both test items (Table 6).

Table 5. Summary of quantitative GMO testing results that were reported in m/m % (top) and cp/cp % (bottom).

Test item	Incorrectly (m/m %)		LOQ (m/m %)		Unsatisfactory z-	
	reported MU ^a (%)	μ^b	$\sigma^{\mathfrak{c}}$	μ	σ	scores (%)
		D	P-356043-5			
Level 1	15/42 (36%)	0.07	0.06	0.16	0.13	1/48 (2%)
Level 2	13/ 42 (30/0)	0.07	0.00	0.10	0.13	1/40 (270)
Test item	Incorrectly		OD cp %)	LC (cp/d)Q cp %)	Unsatisfactory z-
Test item	Incorrectly reported MU (%)					Unsatisfactory z- scores (%)
Test item	<u> </u>	(cp/c	ср %)	(cp/c	cp %)	-
Test item Level 1	<u> </u>	(cp/c	cp %) σ	(cp/c	cp %)	1

^a MU = Measurement Uncertainty

Table 6. Laboratories with outlying z-scores on the basis of the robust mean for Level 1 and 2 test items expressed in m/m % and cp/cp %.

			Unsatisfactory z-scores			
Laboratory	/ Category	ategory Group	m/m%		cp/c	cp%
number	outogory	Огоар	Level 1	Level 2	Level 1	Level 2
L44	(b)	2	Х	Х		
L31	(b)	2			Χ	Χ

 $^{^{}b}$ μ = Sample mean of reported LOD/LOQ values

 $^{^{\}rm c}$ σ = Sample standard deviation of reported LOD/LOQ values

5. Discussion of results

5.1 Overall performance

In this CT round most laboratories correctly identified rice and soybean species in the test items. Through screening for the presence of GM material, 71 out of 85 laboratories reported a positive outcome and most of these identified the correct GM event. GM quantification was also characterised by a satisfactory performance of most laboratories.

All underperforming laboratories for at least one of the four tasks requested in this CT round are listed in Table 7, together with the specification of the failed task (note: this table does not include the labs that did not correctly report the MU).

Of the 27 laboratories that underperformed at least for one task, 8 were NRLs, of which one was appointed under Regulation 882/2004, 5 were appointed under Regulation 1981/2006 and two were appointed under both Regulations. Of the remaining laboratories, 7 were official control laboratories from within the EU and 12 were from third countries.

The common reason of underperformance in the species identification task was the incomplete fulfilment of this task, *i.e.* the labs did not test for the presence of oilseed rape and/or rice. The reason for this incompleteness is not clear, as the species to be screened for were clearly listed in the invitation letter. It may be that some laboratories do not routinely test for these species, and may not have the methods or reagents available to do so. This issue needs further investigation as it suggests that some laboratories do not systematically test all GMOs that have been approved in the EU.

Data on GM detection were lacking for 14 laboratories, yet it is not clear if laboratories not reporting any GM element or GM event did not detect them or did not report them correctly. On GM event identification, over 10% of laboratories reporting the presence of GM material (*i.e.* 8 or 9 depending on the test item) were not able to successfully identify soybean event DP-356043-5 that was present in the samples at relevant levels of 0.7 and 1.5 m/m % GM for Level 1 and 2, respectively. This finding is worrying, because this CT round mimicked real-life samples that may be received by a laboratory and are subjected to GM testing. The underperformance of a number of laboratories in these tasks highlights the need for further guidance in this area.

GM quantification results (for DP-356043-5 soybean), on the other hand, were satisfactory, indicating that most laboratories that provided quantitative data are successful in performing quantitative analyses for this event. The results of two NRLs with z-scores (|z|) \geq 2.0 require further investigation.

Table 7. List of laboratories underperforming for one of the tasks requested and reason for underperformance.

Laboratory number	Category	Group	Species identification	GM detection	GM identification	GM quantification
L35	(a)	1	-	-	Identified maize MON 810 only	Quantitative results for MON 810 only
L05	(a)	3	-	-	-	No quantitative result submitted
L09	(a)	3	-	No GM presence reported	No GM event reported	No quantitative result submitted
L13	(a)	3	No testing for rice	No GM presence reported	No GM event reported	No quantitative result submitted
L25	(b)	2	-	-	-	No quantitative result submitted
L31	(b)	2	-	-	-	z ≥ 2.0 for both test items
L32	(b)	2	-	-	Identified RR soybean only	No quantitative result submitted
L40	(b)	2	-	No GM presence reported	No GM event reported	No quantitative result submitted
L44	(b)	2	<u>-</u>	-	-	z ≥ 2.0 for both test items
L86	(b)	2	No testing for oilseed rape	-	-	-
L06	(c)	5	-	No GM presence reported	No GM event reported	No quantitative result submitted
L14	(c)	5	No testing for rice and oilseed rape	No GM presence reported	No GM event reported	No quantitative result submitted
L36	(c)	5	No testing for oilseed rape	-	No GM event reported	No quantitative result submitted
L43	(c)	5	-	-	-	No quantitative result submitted
L66	(c)	5	-	-	No GM event reported	No quantitative result submitted
L79	(c)	5	-	No GM presence reported	No GM event reported	No quantitative result submitted
L80	(c)	5	No testing for oilseed rape	No GM presence reported	No GM event reported	No quantitative result submitted
L84	(c)	5	No testing for oilseed rape	No GM presence reported	No GM event reported	No quantitative result submitted
L12	(c)	6	-	No GM presence reported	No GM event reported	No quantitative result submitted
L39	(c)	6	No testing for oilseed rape	No GM presence reported	No GM event reported	No quantitative result submitted
L41	(c)	6	Soybean not detected in Level 2; no testing for rice and oilseed rape	No GM presence reported	No GM event reported	No quantitative result submitted
L49	(c)	6	-	No GM presence reported	No GM event reported	No quantitative result submitted
L50	(c)	6	No testing for rice	-	- -	-
L51	(c)	6	-	No GM presence reported	No GM event reported	No quantitative result submitted
L54	(c)	6	-	No GM presence reported	No GM event reported	No quantitative result submitted
L63	(c)	6	-	-	No GM event reported	No quantitative result submitted
L67	(c)	6	No testing for oilseed rape	-	-	-
L69	(c)	6	No testing for oilseed rape	-	No GM event reported	No quantitative result submitted
L82	(c)	6	No testing for rice and oilseed rape	-	No GM event reported	No quantitative result submitted
L83	(c)	6	No testing for oilseed rape	=	Identified soybean 40-	No quantitative result submitted

[&]quot;-" means no underperformance for a task was observed.

5.2 Measurement uncertainty

In Figure 5 the robust means (μ_R) for soybean event DP-356043-5, expressed in m/m % and cp/cp %, are shown, and the expanded measurement uncertainties (see Table 4) are indicated by vertical bars.

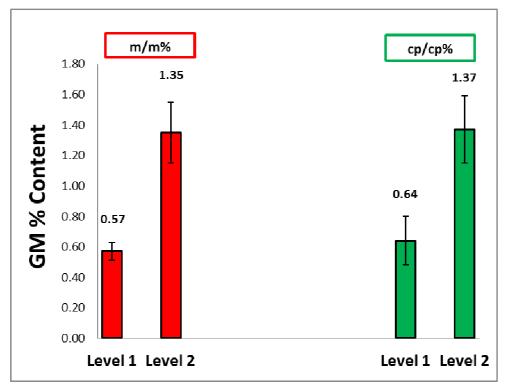


Figure 5. Robust means (μ_R) and measurement uncertainties (vertical bars) of Level 1 and 2 test items for soybean event DP-356043-5.

It is worth noting that on average, across test items, only 33 out of 57 laboratories that provided quantitative results for DP-356043-5 soybean (approximately 58 %) reported a complete and consistent estimate of the measurement uncertainty (MU), a trend in reporting MU which is better than in the previous CT round. In particular:

- 12 laboratories (approximately 21 %) answered the questions relating to MU on the questionnaire inconsistently, meaning that they answered yes to both question 35.1. and 35.2, which is clearly not possible because the two questions are mutually exclusive;
- 3 laboratories (approximately 5 %) did not answer all questions relating to MU;
- 6 laboratories (approximately 11 %) did not provide any estimate of the MU;
- 4 laboratories (approximately 7 %) reported a relative estimate, even though the questionnaire explicitly stated that an absolute value had to be reported for the MU.

6. Conclusions

Participants in this CT round were required to analyse two test items consisting of a mixture of rice noodles and soybean event DP-356043-5 at different concentrations (Level 1 and 2 test items), *i.e.*:

- Perform species identification: maize, soybean, oilseed rape and rice
- Identify and quantify the GM event(s) detected.

Laboratories' performance, with respect to the different tasks to be performed, has been overall satisfactory, with some exceptions. The following general conclusions could be drawn:

- Despite the fact that almost all laboratories correctly detected soybean and rice, there were
 few that did not test for all species, though explicitly requested. The reason for this deficiency
 has to be investigated. In principle, every laboratory should be able to screen for all the
 common plant species, as real-life samples generally come without any a priori information on
 the sample matrix.
- A failure in GM detection or reporting of such data was noted for a number of laboratories and further follow-up is needed, *e.g.* by requesting additional information to the laboratories and addressing any problems reported.
- The failure in the identification of soybean event DP-356043-5 by a number of laboratories suggests that improvements are needed in this area, as in principle all laboratories should be able to identify an event for which a validated method exists, without possessing any information on the event(s) actually present in the samples.
- Most laboratories that quantified soybean event DP-356043-5 performed satisfactorily on this task.

In general, it is important that the competence of a laboratory to fulfil its mandate is measured not only by evaluating z-scores on the quantitative results obtained, but by investigating the whole analytical approach from species identification, GM detection and identification, and finally GM event quantification. Such comprehensive evaluation allows identifying issues and limitations in current practices and should on the long term improve and harmonise the performance of GMO testing laboratories.

The measurement uncertainty (MU) was, on average across concentration levels, reported in a complete and consistent manner by approximately 58 % of laboratories, a result which is better than the one obtained in the previous CT round (46 %, p-value > 0.05 with Normal approximation test). Despite this improvement, and given the importance of a correct estimation of the measurement uncertainty, there is still a need to provide laboratories with guidance and training on this topic.

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The laboratories listed below are kindly acknowledged for their participation in this exercise.

Organisation	Department	Country Code	Group
AGES - Institute for Food Safety Vienna		AT	3
Agricultural Genetics Institute	GMO Detection Laboratory	VN	6
Agricultural Institute of Slovenia		SL	2
Agri-Food and Veterinary Authority of Singapore	Laboratories Group	SG	6
Agroscope, Institute for Livestock Sciences ILS	Feed Biology	СН	6
American University of Science and Technology	Laboratory Science &Technology	LB	6
ASL Milano 1	Laboratorio DI Prevenzione	IT	5
Bavarian Health and Food Safety Authority (LGL)		DE	2
BIOMI LTD		HU	4
Bundesamt für Verbraucherschutz und Lebensmittelsicherheit		DE	1
Centre wallon de recherches agronomiques	Valorisation des productions	BE	3
Centro Nacional de Alimentacion (Agencia Espanola de Consumo Seguridad Alimentaria y Nutricion)	Biotechnology Unit	ES	3
Chemischen und Veterinäruntersuchungsamt Rhein-Ruhr-Wupper (CVUA-RRW)	FG 40-5	DE	4
CRA-SCS	sede di Tavazzano- Laboratorio	ΙΤ	2
Croatian centre for agriculture, food and rural affairs, Institute for seed and seedlings	Seed testing laboratory	HR	4
Crop Research Institute		CZ	3
CVUA Freiburg	GMO Testing	DE	2
Danish Veterinary and Food Agency	Plantdiagnostics	DK	3
ERSA	Servizio fitosanitario	IT	5
FASFC	FASFC Melle	BE	5
Federal Food Safety and Veterinary Office FSVO	Risk Assessment Division	СН	4
Fera		UK	2
Finnish Customs Laboratory		FI	3
Finnish Food Safety Authority Evira	Research and Laboratory Dept	FI	2
ILVO	Technology & Food Sciences	BE	3
INIAV		PT	3
Institut für Hygiene und Umwelt	Gentechnik	DE	2
Institute for Animal Health, Food Safety and Environment	Virology	LV	3
Institute for Diagnosis and Animal Health	Molecular Biology and GMO	RO	1
Institute of Biochemistry and Biophysics PAS		PL	2
Instytut Zootechniki PIB KLP Pracownia w Szczecinie		PL	3
Istituto Zooprofilattico Sperimentale del Piemonte, Liguria e Valle d'Aosta		IT	5
Istituto Zooprofilattico Sperimentale della Lombardia ed Emilia Romagna	Reparto Genomica	IT	5
Istituto Zooprofilattico Sperimentale della Sardegna	Igiene degli Alimenti- Control	IT	5
Istituto Zooprofilattico Sperimentale dell'Abruzzo e del Molise G. Caporale	Food Hygiene	IT	5

Organisation	Department	Country Code	Group
Istituto Zooprofilattico Sperimentale delle Regioni Lazio e Toscana	Biotecnology Unit	IT	3
Istituto Zooprofilattico Sperimentale delle Venezie	Department of food safety	IT	5
Laboratoire National de Santé	Food Control	LU	3
Laboratorio Arbitral Agroalimentario, LAA-MAGRAMA	OGM	ES	3
Laboratory of SGS Bulgaria Ltd		BG	5
LANAGRO-MG	PRIMAR	BR	6
Landesamt für Landwirtschaft, Lebensmittelsicherheit und Fischerei Mecklenburg-Vorpommern		DE	2
Landesamt für Verbraucherschutz Sachsen-Anhalt	Dezermnat 31	DE	2
Landesbetrieb Hessisches Landeslabor		DE	2
Landeslabor Berlin-Brandenburg	Fb I-6	DE	2
Landeslabor Schleswig-Holstein	Institut f.	DE	2
Landesuntersuchungsamt Rheinland-Pfalz	Lebensmittelchemie	DE	2
Landesuntersuchungsanstalt für das Gesundheits- und Veterinärwesen Sachsen (LUA)	Amtliche Lebensmitteluntersuch	DE	2
LAV Landesamt f Verbraucherschutz		DE	2
LAVES LVI Braunschweig/Hannover	FB 12	DE	2
LGC	Molecular Biology (Block 7)	UK	3
LTZ Augustenberg	(======)	DE	2
Ministry of Agriculture Livestock an Food Supply	LANAGRO-GO - LDV	BR	6
Ministry of Finance, General Chemical State Laboratory	Food Directorate	GR	3
Ministry of Food-Agriculture and livestock	Provincial Control Laboratory	TR	6
National Bureau of Plant Genetic Resources	Genomic Resources	IN	6
National Center for Molecular Characterization of Genetically Modified	Division School of Life Sciences	CN	6
Organisms, SJTU	& Bio		
National Center of Public Health and Analyses	GMO Unit	BG SE	3
National Food Agency	Science department Molecular Biology and		
National Food and Veterinary Risk Assesment Institute	GMO	LT	3
National Food Chain Safety Office	Biotechnology & GMO	HU	3
National Food Reference Laboratory	Unit	TR	6
National Institute of Biology		SI	3
National institute of public health in Prague		CZ	2
National Public Health Laboratory	Food	MY	6
National Veterinary Research Institute		PL	3
Netherlands Food and Consumer Product Safety Authority(NVWA)	Laboratorium VV Lab. Biotechnology	NL	2
Nstional Agency of Drug and Food Control / BADAN POM RI	PPOMN	ID	6
Quality assurance and Testing centre 3	Microbiology - GMO testing lab	VN	6
Regional Laboratory of Genetically Modified Food		PL	3
RIKILT Wageningen UR	NFA	NL	3
Scientific Institute of Public Health	PBB	BE	3
Service Commun des Laboratoires		BE	3
Servicio Agricola y Ganadero	De laboratorios y Estaciones C	CL	6
SP Laboratorija A.D. BEČEJ		RS	6
Staatliche Betriebsgesellschaft für Umwelt und Landwirtschaft	Geschäftsbereich 6, FB 63	DE	2
State General Laboratory		CY	3
State Veterinary and Food Institute, VFI in Dolný Kubín		SK	3
Tallinn University of Technology	Department of Gene Technology	EE	2
Thüringer Landesamt für Verbraucherschutz	Lab for detection of GMO/foods	DE	2
Thüringer Landesanstalt für Landwirtschaft	Untersuchungswesen	DE	4
Ukrmetrteststandard	Scientific-Resrarch	UA	6
Umweltbundesamt GmbH	Center	AT	3
	Haematology and Cell	ZA	6
University of the Free State	Biology/G		

¹ See laboratory groups description on page 6.

Annex 1: Homogeneity and stability of test items

A1.1 Homogeneity of test items

The assessment of the homogeneity⁽¹¹⁾ was performed by the EU-RL GMFF after the test items had been packed in their final form and before distribution to participants, using the following acceptance criterion:

$$s_s \le 0.3 \overset{\circ}{\sigma}$$
 (A1.1)

Where s_{s} is the between-test item standard deviation as determined by a 1-way random effects

ANOVA⁽¹²⁾ and $\overset{\wedge}{\sigma}$ is the standard deviation for comparative testing. The value of $\overset{\wedge}{\sigma}$, the target standard deviation for comparative testing, was defined by the Members of the Advisory Board on the basis of the experience acquired with previous CT rounds, and set to $0.2^{(13)}$.

If the criterion according to A1.1 is met, the between-test item standard deviation contributes no more than about 10 % to the standard deviation for comparative testing.

The repeatability of the test method is the square root of the mean sum of squares within-test items MS_{within} . The relative between-test item standard deviation $s_{s,rel}$ is given by

$$s_{s,rel} = \frac{\sqrt{\frac{MS_{between} - MS_{within}}{n}}}{\frac{n}{\overline{v}}} \times 100\%$$
 (A1.2)

where: $MS_{between}$ is the mean sum of squares between test items

MSwithin is the mean sum of squares within test items

n is the number of replicates for each sample

 \overline{y} is the mean of the homogeneity data

If $MS_{within} > MS_{between}$, then:

$$s_{s,rel} = u_{bb}^* = \frac{\frac{repeatability}{\sqrt{n}} \sqrt[4]{\frac{2}{N(n-1)}} \times 100\%$$
 (A1.3)

where: u^*_{bb} is the maximum uncertainty contribution that can be obtained by the hidden heterogeneity of the material.

For each group of test items 10 bottles (N = 10) were randomly selected and analysed in five-fold replicates (n = 5). The criterion described in formula (A1.1) was in all cases fulfilled, indicating that all groups of test items were homogeneous. The data from the homogeneity study were also used for the estimation of the uncertainty contribution related to the level of homogeneity of test items.

A1.2 Stability of test items

An isochronous short term stability study involving two Level 1 test samples with three replicates each (N = 2, n = 3), was conducted over one, two and four weeks at $+4^{\circ}$ C and $+18^{\circ}$ C (14). The results did not reveal an influence of time or temperature on the stability of the test items. The test items were therefore shipped at ambient temperature. Within the time period of this comparative study, the test materials were considered sufficiently stable.

Annex 2: Species identification

A2.1 Rice

Overall, 100 % and 98 % of laboratories that tested for the presence of rice species for Level 1 and 2 test items, respectively, identified rice in the samples, without any difference between laboratory categories. The two laboratories that did not detect this species in the Level 2 test item while detecting it in Level 1 appears to have inadvertently indicated "absence" for the former test item for all species, an error that might have been due to the fact that the order of the first two alternative answers ("Absence" and "Presence") was reversed in the questionnaire between Level 1 and Level 2 questions.

Table A2.1. Results of species identification analysis for rice by laboratory category; D = Detected, ND = Not Detected. When estimating overall percentages (D and ND) per category, only laboratories that actually tested for rice were considered in the denominator.

(a)						
LabCode	Group	Level 1	Level 2			
L35	1	Detected	Detected			
L53	1	Detected	Detected			
L01	3	Detected	Detected			
L03	3	Detected	Detected			
L04	3	Detected	Detected			
L05	3	Detected	Detected			
L08	3	Detected	Detected			
L09	3	Detected	Detected			
L11	3	Detected	Detected			
L13	3	Not Tested	Not Tested			
L16	3	Detected	Detected			
L18	3	Detected	Detected			
L19	3	Detected	Detected			
L21	3	Detected	Detected			
L25	3	Detected	Detected			
L26	3	Detected	Detected			
L34	3	Detected	Detected			
L42	3	Detected	Detected			
L45	3	Detected	Detected			
L56	3	Detected	Detected			
L58	3	Detected	Detected			
L59	3	Detected	Detected			
L60	3	Detected	Detected			
L61	3	Detected	Detected			
L64	3	Detected	Detected			
L65	3	Detected	Detected			
L70	3	Detected	Detected			
L72	3	Detected	Detected			
L85	3	Detected	Detected			
L88	3	Detected	Detected			
	% D	100	100			

% ND

(b)					
LabCode	Group	Level 1	Level 2		
L17	2	Detected	Detected		
L20	2	Detected	Detected		
L22	2	Detected	Detected		
L23	2	Detected	Detected		
L24	2	Detected	Detected		
L27	2	Detected	Detected		
L28	2	Detected	Detected		
L29	2	Detected	Detected		
L31	2	Detected	Detected		
L32	2	Detected	Not Detected		
L33	2	Detected	Detected		
L37	2	Detected	Detected		
L40	2	Detected	Detected		
L44	2	Detected	Detected		
L48	2	Detected	Detected		
L52	2	Detected	Detected		
L62	2	Detected	Detected		
L71	2	Detected	Detected		
L74	2	Detected	Detected		
L75	2	Detected	Detected		
L78	2	Detected	Detected		
L86	2	Detected	Detected		
L87	2	Detected	Detected		
	% D	100	96		
	% ND	0	4		

Left			(c)	
L55 4 Detected Detected L68 4 Detected Detected L76 4 Detected Detected L81 4 Detected Detected L81 4 Detected Detected L06 5 Detected Detected L14 5 Not Tested Not Tested L30 5 Detected Detected L33 5 Detected Detected L43 5 Detected Detected L43 5 Detected Detected L44 5 Detected Detected L49 5 Detected Detected L402 6 Detected Detected L403 6 Detected Detected L404 6 Detected Detected L407 6 Detected Detected L41 6 Not Tested Not Tested L47	LabCode	Group	Level 1	Level 2
L68 4 Detected Detected L76 4 Detected Detected L81 4 Detected Detected L81 4 Detected Detected L06 5 Detected Detected L14 5 Not Tested Not Tested L30 5 Detected Detected L33 5 Detected Detected L43 5 Detected Detected L66 5 Detected Detected L80 5 Detected Detected L81 5 Detected Detected L82 6 Detected Detected L81 6 Detected Detected L82	L46	4	Detected	Detected
LT6 4 Detected Detected LB1 4 Detected Detected LL6 5 Detected Detected LL14 5 Not Tested Not Tested LL30 5 Detected Detected LL36 5 Detected Detected LL33 5 Detected Detected LL66 5 Detected Detected LL79 5 Detected Detected LL80 6 Detected Detected LL80 6 Detected Detected LL02 6 Detected Detected Detected LL10 6 Detected Detected Detected LL12 6 Detected <td< td=""><td>L55</td><td>4</td><td>Detected</td><td>Detected</td></td<>	L55	4	Detected	Detected
L81 4 Detected Detected L06 5 Detected Detected L14 5 Not Tested Not Tested L30 5 Detected Detected L36 5 Detected Detected L43 5 Detected Detected L66 5 Detected Detected L79 5 Detected Detected L80 5 Detected Detected L84 5 Detected Detected L84 5 Detected Detected L02 6 Detected Detected L07 6 Detected Detected L10 6 Detected Detected L10 6 Detected Detected L12 6 Detected Detected L23 6 Detected Detected L41 6 Not Tested Not Tested L50	L68	4	Detected	Detected
LO6 5 Detected Detected L14 5 Not Tested Not Tested L30 5 Detected Detected L36 5 Detected Detected L43 5 Detected Detected L66 5 Detected Detected L48 5 Detected Detected L80 5 Detected Detected L84 5 Detected Detected L02 6 Detected Not Detected Detected L07 6 Detected Detected Detected Detected L10 6 Detected Detected	L76	4	Detected	Detected
L14 5 Not Tested Not Tested L30 5 Detected Detected L36 5 Detected Detected L43 5 Detected Detected L66 5 Detected Detected L29 5 Detected Detected L80 5 Detected Detected L84 5 Detected Detected L02 6 Detected Not Detected Detected L07 6 Detected Detected Detected L10 6 Detected Detected Detected L12 6 Detected Detected Detected L39 6 Detected Detected Detected L41 6 Not Tested Not Tested Not Tested L47 6 Detected Detected Detected L50 6 Not Tested Not Tested Detected Detected	L81	4	Detected	Detected
L30 5 Detected Detected L36 5 Detected Detected L43 5 Detected Detected L66 5 Detected Detected L29 5 Detected Detected L80 5 Detected Detected L84 5 Detected Detected L02 6 Detected Not Detected L07 6 Detected Detected L10 6 Detected Detected L10 6 Detected Detected L12 6 Detected Detected L39 6 Detected Detected L41 6 Not Tested Not Tested L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54	L06	5	Detected	Detected
L36 5 Detected Detected L43 5 Detected Detected L66 5 Detected Detected L29 5 Detected Detected L80 5 Detected Detected L84 5 Detected Detected L02 6 Detected Not Detected L07 6 Detected Detected L10 6 Detected Detected L10 6 Detected Detected L12 6 Detected Detected L39 6 Detected Detected L41 6 Not Tested Not Tested L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L51 6 Detected Detected L63	L14	5	Not Tested	Not Tested
L43 5 Detected Detected L66 5 Detected Detected L29 5 Detected Detected L80 5 Detected Detected L84 5 Detected Detected L02 6 Detected Detected L07 6 Detected Detected L10 6 Detected Detected L12 6 Detected Detected L39 6 Detected Detected L41 6 Not Tested Not Tested L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69	L30	5	Detected	Detected
L66 5 Detected Detected L79 5 Detected Detected L80 5 Detected Detected L84 5 Detected Detected L02 6 Detected Not Detected L07 6 Detected Detected L10 6 Detected Detected L12 6 Detected Detected L39 6 Detected Detected L41 6 Not Tested Not Tested L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73	L36	5	Detected	Detected
LT9 5 Detected Detected L80 5 Detected Detected L84 5 Detected Detected L02 6 Detected Not Detected L07 6 Detected Detected L10 6 Detected Detected L12 6 Detected Detected L39 6 Detected Detected L41 6 Not Tested Not Tested L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77	L43	5	Detected	Detected
L80 5 Detected Detected L84 5 Detected Detected L02 6 Detected Not Detected L07 6 Detected Detected L10 6 Detected Detected L12 6 Detected Detected L39 6 Detected Detected L41 6 Not Tested Not Tested L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L77	L66	5	Detected	Detected
L84 5 Detected Detected L02 6 Detected Not Detected L07 6 Detected Detected L10 6 Detected Detected L12 6 Detected Detected L39 6 Detected Detected L41 6 Not Tested Not Tested L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L82 6 Not Tested Not Tested	L79	5	Detected	Detected
LO2 6 Detected Not Detected LO7 6 Detected Detected L10 6 Detected Detected L12 6 Detected Detected L39 6 Detected Detected L41 6 Not Tested Not Tested L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L82 6 Not Tested Not Tested	L80	5	Detected	Detected
LO7 6 Detected Detected L10 6 Detected Detected L12 6 Detected Detected L39 6 Detected Detected L41 6 Not Tested Not Tested L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77	L84	5	Detected Detected	
L10 6 Detected Detected L12 6 Detected Detected L39 6 Detected Detected L41 6 Not Tested Not Tested L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L77 6 Detected Not Tested L82 6 Not Tested Detected	L02	6	Detected	Not Detected
L12 6 Detected Detected L39 6 Detected Detected L41 6 Not Tested Not Tested L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L77 6 Detected Not Tested L82 6 Not Tested Detected	L07	6	Detected	Detected
L39 6 Detected Detected L41 6 Not Tested Not Tested L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L82 6 Not Tested Not Tested L83 6 Detected Detected	L10	6	Detected	Detected
L41 6 Not Tested Not Tested L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L82 6 Not Tested Not Tested L83 6 Detected Detected	L12	6	Detected	Detected
L47 6 Detected Detected L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L82 6 Not Tested Not Tested L83 6 Detected Detected	L39	6	Detected	Detected
L49 6 Detected Detected L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L82 6 Not Tested Not Tested L83 6 Detected Detected	L41	6	Not Tested	Not Tested
L50 6 Not Tested Not Tested L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L82 6 Not Tested Not Tested L83 6 Detected Detected	L47	6	Detected	Detected
L51 6 Detected Detected L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L82 6 Not Tested Not Tested L83 6 Detected Detected	L49	6	Detected	Detected
L54 6 Detected Detected L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L82 6 Not Tested Not Tested L83 6 Detected Detected	L50	6	Not Tested	Not Tested
L63 6 Detected Detected L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L82 6 Not Tested Not Tested L83 6 Detected Detected	L51	6	Detected	Detected
L67 6 Detected Detected L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L82 6 Not Tested Not Tested L83 6 Detected Detected	L54	6	Detected	Detected
L69 6 Detected Detected L73 6 Detected Detected L77 6 Detected Detected L82 6 Not Tested Not Tested L83 6 Detected Detected	L63	6	Detected	Detected
L73 6 Detected Detected L77 6 Detected Detected L82 6 Not Tested Not Tested L83 6 Detected Detected	L67	6	Detected	Detected
L77 6 Detected Detected L82 6 Not Tested Not Tested L83 6 Detected Detected	L69	6	Detected	Detected
L82 6 Not Tested Not Tested L83 6 Detected Detected	L73	6	Detected	Detected
L83 6 Detected Detected	L77	6	Detected	Detected
· · · · · · · · · · · · · · · · · · ·	L82	6	Not Tested	Not Tested
% D 100 96	L83	6	Detected	Detected
		% D	100	96

EU-RL GMFF: Comparative testing report

A2.2 Soybean

All 85 laboratories that screened for this species correctly detected it in the Level 1 test item whereas for Level 2 81 laboratories (approximately 94 %) reported the presence of soybean in the samples. In 4 out of 5 cases this is likely due to an error in the filling of the questionnaire rather than to an actual error of identification; however, one laboratory acting as official control laboratory from outside the EU actually identified soybean only in the Level 1 test item.

Table A2.2. Results of species identification analysis for soybean by laboratory category; D = Detected, ND = Not Detected. When estimating overall percentages (D and ND) per category, only laboratories that actually tested for soybean were considered in the denominator.

		(a)	
LabCode	Group	Level 1	Level 2
L35	1	Detected	Detected
L53	1	Detected	Detected
L01	3	Detected	Detected
L03	3	Detected	Detected
L04	3	Detected	Detected
L05	3	Detected	Detected
L08	3	Detected	Detected
L09	3	Detected	Detected
L11	3	Detected	Detected
L13	3	Detected	Detected
L16	3	Detected	Detected
L18	3	Detected	Detected
L19	3	Detected	Detected
L21	3	Detected	Detected
L25	2	Detected	Detected
L26	3	Detected	Detected
L34	3	Detected	Detected
L42	3	Detected	Detected
L45	3	Detected	Detected
L56	3	Detected	Detected
L58	3	Detected	Detected
L59	3	Detected	Detected
L60	3	Detected	Detected
L61	3	Detected	Detected
L64	3	Detected	Detected
L65	3	Detected	Detected
L70	3	Detected	Detected
L72	3	Detected	Detected
L85	3	Detected	Detected
L88	3	Detected	Detected
	% D	100	100

% ND

		(b)	
LabCode	Group	Level 1	Level 2
L17	2	Detected	Detected
L20	2	Detected	Detected
L22	2	Detected	Detected
L23	2	Detected	Detected
L24	2	Detected	Detected
L27	2	Detected	Detected
L28	2	Detected	Detected
L29	2	Detected	Detected
L31	2	Detected	Detected
L32	2	Detected	Not Detected
L33	2	Detected	Detected
L37	2	Detected	Detected
L40	2	Detected	Detected
L44	2	Detected	Detected
L48	2	Detected	Detected
L52	2	Detected	Detected
L62	2	Detected	Detected
L71	2	Detected	Detected
L74	2	Detected	Detected
L75	2	Detected	Detected
L78	2	Detected	Detected
L86	2	Detected	Detected
L87	2	Detected	Detected
	% D	100	96
	% ND	0	4

		(c)	
LabCode	Group	Level 1	Level 2
L46	4	Detected	Detected
L55	4	Detected	Detected
L68	4	Detected	Detected
L76	6	Detected	Detected
L81	4	Detected	Detected
L06	5	Detected	Detected
L14	5	Detected	Detected
L30	5	Detected	Detected
L36	5	Detected	Detected
L43	5	Detected	Detected
L66	5	Detected	Detected
L79	5	Detected	Detected
L80	5	Detected	Detected
L84	5	Detected	Detected
L02	6	Detected	Not Detected
L07	6	Detected	Detected
L10	6	Detected	Detected
L12	6	Detected	Detected
L39	6	Detected	Detected
L41	6	Detected	Not Detected
L47	6	Detected	Detected
L49	6	Detected	Detected
L50	6	Detected	Detected
L51	6	Detected	Detected
L54	6	Detected	Detected
L63	6	Detected	Detected
L67	6	Detected	Detected
L69	6	Detected	Detected
L73	6	Detected	Detected
L77	6	Detected	Detected
L82	6	Detected	Not Detected
L83	6	Detected	Detected
	% D	100	91
		_	_

A2.3 Maize

Also in this case inversion of answers presumably resulted in differences between the results reported for Level 1 and Level 2 test items, with respectively 90 % and 87 % of laboratories correctly not detecting maize species. A few laboratories did detect maize.

Table A2.3. Results of species identification analysis for maize by laboratory category; D = Detected, ND = Not Detected. When estimating overall percentages (D and ND) per category, only laboratories that actually tested for maize were considered in the denominator.

		(a)	
LabCode	Group	Level 1	Level 2
L35	1	Detected	Detected
L53	1	Not Detected	Not Detected
L01	3	Not Detected	Not Detected
L03	3	Not Detected	Not Detected
L04	3	Not Detected	Not Detected
L05	3	Not Detected	Not Detected
L08	3	Detected	Detected
L09	3	Not Detected	Not Detected
L11	3	Not Detected	Not Detected
L13	3	Detected	Detected
L16	3	Not Detected	Not Detected
L18	3	Not Detected	Not Detected
L19	3	Not Detected	Not Detected
L21	3	Not Detected	Not Detected
L25	3	Not Detected	Not Detected
L26	3	Not Detected	Not Detected
L34	3	Not Detected	Not Detected
L42	3	Not Detected	Not Detected
L45	3	Not Detected	Not Detected
L56	3	Not Detected	Not Detected
L58	3	Not Detected	Not Detected
L59	3	Not Detected	Not Detected
L60	3	Not Detected	Not Detected
L61	3	Not Detected	Not Detected
L64	3	Not Detected	Not Detected
L65	3	Not Detected	Not Detected
L70	3	Not Detected	Not Detected
L72	3	Not Detected	Not Detected
L85	3	Not Detected	Not Detected
L88	3	Not Detected	Not Detected
	% D	10	10

% ND

		(b)	
LabCode	Group	Level 1	Level 2
L17	2	Not Detected	Not Detected
L20	2	Not Detected	Not Detected
L22	2	Detected	Detected
L23	2	Not Detected	Not Detected
L24	2	Not Detected	Not Detected
L27	2	Not Detected	Not Detected
L28	2	Not Detected	Not Detected
L29	2	Not Detected	Not Detected
L31	2	Not Detected	Not Detected
L32	2	Not Detected	Detected
L33	2	Not Detected	Not Detected
L37	2	Not Detected	Not Detected
L40	2	Detected	Detected
L44	2	Not Detected	Not Detected
L48	2	Not Detected	Not Detected
L52	2	Not Detected	Not Detected
L62	2	Not Detected	Not Detected
L71	2	Not Detected	Not Detected
L74	2	Not Detected	Not Detected
L75	2	Not Detected	Not Detected
L78	2	Not Detected	Not Detected
L86	2	Not Detected	Not Detected
L87	2	Not Detected	Not Detected
	% D	9	13

% ND

(c)				
LabCode	Group	Level 1	Level 2	
L46	4	Not Detected	Not Detected	
L55	4	Not Detected	Not Detected	
L68	4	Not Detected	Not Detected	
L76	6	Not Detected	Not Detected	
L81	4	Not Detected	Not Detected	
L06	5	Not Detected	Not Detected	
L14	5	Not Detected	Not Detected	
L30	5	Not Detected	Not Detected	
L36	5	Not Detected	Not Detected	
L43	5	Detected	Detected	
L66	5	Not Detected	Not Detected	
L79	5	Not Detected	Not Detected	
L80	5	Not Detected	Not Detected	
L84	5	Not Detected	Not Detected	
L02	6	Not Detected	Detected	
L07	6	Not Detected	Not Detected	
L10	6	Not Detected	Not Detected	
L12	6	Not Detected	Not Detected	
L39	6	Not Detected	Not Detected	
L41	6	Detected	Detected	
L47	6	Not Detected	Not Detected	
L49	6	Not Detected	Not Detected	
L50	6	Not Detected	Not Detected	
L51	6	Not Detected	Not Detected	
L54	6	Not Detected	Not Detected	
L63	6	Not Detected	Not Detected	
L67	6	Detected	Detected	
L69	6	Not Detected	Not Detected	
L73	6	Not Detected	Not Detected	
L77	6	Not Detected	Not Detected	
L82	6	Not Detected	Detected	
L83	6	Not Detected	Not Detected	
	% D	9	16	
	% ND	91	84	

A2.4 Oilseed Rape

Overall 99 % and 95 % of laboratories who tested for the presence of oilseed rape, for Level 1 and Level 2 respectively, did not detect it. While three laboratories likely "swapped" their answer for Level 2, one laboratory in category (c) identified oilseed rape in both test items. Out of 85 laboratories, however, only 74 (approximately 87 %) actually tested for this species, and most of those that did not test belonged to category (c), with the exception of two NRLs, one appointed under both Regulations and one under Regulation 1981/2006 only.

Table A2.4. Results of species identification analysis for oilseed rape by laboratory category; D = Detected, ND = Not Detected. When estimating overall percentages (D and ND) per category, only laboratories that actually tested for oilseed rape were considered in the denominator.

		(a)	
LabCode	Group	Level 1	Level 2
L35	1	Not Detected	Not Detected
L53	1	Not Detected	Not Detected
L01	3	Not Detected	Not Detected
L03	3	Not Detected	Not Detected
L04	3	Not Detected	Not Detected
L05	3	Not Detected	Not Detected
L08	3	Not Detected	Not Detected
L09	3	Not Detected	Not Detected
L11	3	Not Detected	Not Detected
L13	3	Not Detected	Not Detected
L16	3	Not Detected	Not Detected
L18	3	Not Detected	Not Detected
L19	3	Not Detected	Not Detected
L21	3	Not Detected	Not Detected
L25	2	Not Detected	Not Detected
L26	3	Not Detected	Not Detected
L34	3	Not Detected	Not Detected
L42	3	Not Detected	Not Detected
L45	3	Not Detected	Not Detected
L56	3	Not Detected	Not Detected
L58	3	Not Detected	Not Detected
L59	3	Not Detected	Not Detected
L60	3	Not Detected	Not Detected
L61	3	Not Detected	Not Detected
L64	3	Not Detected	Not Detected
L65	3	Not Detected	Not Detected
L70	3	Not Detected	Not Detected
L72	3	Not Detected	Not Detected
L85	3	Not Detected	Not Detected
L88	3	Not Detected	Not Detected
	% D	0	0

% ND

100

100

		(b)	
LabCode	Group	Level 1	Level 2
L17	2	Not Detected	Not Detected
L20	2	Not Detected	Not Detected
L22	2	Not Detected	Not Detected
L23	2	Not Detected	Not Detected
L24	2	Not Detected	Not Detected
L27	2	Not Detected	Not Detected
L28	2	Not Detected	Not Detected
L29	2	Not Detected	Not Detected
L31	2	Not Detected	Not Detected
L32	2	Not Detected	Detected
L33	2	Not Detected	Not Detected
L37	2	Not Detected	Not Detected
L40	2	Not Detected	Detected
L48	2	Not Detected	Not Detected
L52	2	Not Detected	Not Detected
L62	2	Not Detected	Not Detected
L71	2	Not Detected	Not Detected
L74	2	Not Detected	Not Detected
L75	2	Not Detected	Not Detected
L78	2	Not Detected	Not Detected
L86	2	Not Tested	Not Tested
L87	2	Not Detected	Not Detected
	% D	0	10
	% ND	100	90

(c)				
LabCode	Group	Level 1	Level 2	
L46	4	Not Detected	Not Detected	
L55	4	Not Detected	Not Detected	
L68	4	Not Detected	Not Detected	
L76	6	Not Detected	Not Detected	
L81	4	Not Detected	Not Detected	
L06	5	Not Detected	Not Detected	
L14	5	Not Tested	Not Tested	
L30	5	Not Detected	Not Detected	
L36	5	Not Tested	Not Tested	
L43	5	Not Detected	Not Detected	
L66	5	Not Detected	Not Detected	
L79	5	Not Detected	Not Detected	
L80	5	Not Tested	Not Tested	
L84	5	Not Tested	Not Tested	
L02	6	Not Detected	Detected	
L07	6	Not Detected Not Detected		
L10	6	Not Detected	Not Detected	
L12	6	Not Detected	Not Detected	
L39	6	Not Tested	Not Tested	
L41	6	Not Tested	Not Tested	
L47	6	Not Detected	Not Detected	
L49	6	Not Detected	Not Detected	
L50	6	Not Detected	Not Detected	
L51	6	Not Detected	Not Detected	
L54	6	Not Detected	Not Detected	
L63	6	Not Detected	Not Detected	
L67	6	Not Tested	Not Tested	
L69	6	Not Tested	Not Tested	
L73	6	Detected	Detected	
L77	6	Not Detected	Not Detected	
L82	6	Not Tested	Not Tested	
L83	6	Not Tested	Not Tested	
-	% D	5	9	
	% ND	95	91	

Annex 3: Performance statistics

The aim of performance statistics is to provide participants with a meaningful result that can be easily interpreted. The procedure followed for the evaluation of participants' performance was agreed by the Members of the Advisory Board and relies on the calculation of z-scores from \log_{10} -transformed data^(15,16) based on the robust means^(7,8) (μ_R) of the participants' results.

The EU-RL GMFF calculated the consensus values from participants taking the robust means (μ_R) for Level 1 and 2 test items in m/m % and cp/cp % on both original and \log_{10} -transformed scale, taking into account the agreed standard deviation ($\overset{\circ}{\sigma}$) for comparative testing (see Annex 1).

The z-scores (z_i) for participant i reporting measurement result x_i are calculated in comparison to the robust mean as follows:

$$z_i = (\log_{10} x_i - \log_{10} \mu_R) / \hat{\sigma}$$
 (A3.1)

Annex 4: Participants' results

The z-scores of all laboratories are reported in Tables A4.1-A4.4. For consistency, all decimal numbers were rounded to two digits. The information is given, when the sample size allowed it, by laboratory category and, for indicative purposes, by laboratory group (see page 6). "Value" refers to the reported value and uncertainty as calculated and reported by the laboratory. Also "LOD" (limit of detection) and "LOQ" (limit of quantification) are values calculated and provided by the laboratories and refer to the methods they used for these specific samples. The z-scores, measurement uncertainty (MU; % of incorrectly reported MU is estimated only using data from laboratories which reported a value), mean LOD (μ_{LOD}) and mean LOQ (μ_{LOQ}) as well as their standard deviation are calculated by the EU-RL. As an indicator for the overall performance, the fraction of laboratories outside the acceptable range of the z-score is given and corresponding data are highlighted in bold.

Table A4.1a-c. z-scores for soybean event DP-356043-5 **Level 1 test item** for results reported in m/m %, laboratory category: (a), (b), and (c). -= not reported, (1) Uncertainty (U) and/or coverage factor K was reported in an inconsistent manner, (2) U was reported in an incomplete manner, (3) U seems to be a relative value. Descriptive statistics for LOD and LOQ were estimated using only data for which a value was clearly reported.

(a)

Laboratory Number	Group	Value	Uncertainty	LOD m/m	LOQ m/m	z-score based on $\mu_R = 0.57$
L53	1	0.72	0.20	0.03	0.11	0.55
L01	3	0.56	0.22	-	-	0.01
L03	3	0.54	0.10	-	-	-0.06
L04	3	0.71	69.10 (3)	-	-	0.53
L08	3	0.47	30.00 (3)	0.09	-	-0.37
L11	3	0.40	0.02 (1)	-	-	-0.72
L16	3	0.33	0.12 (1)	0.03	0.10	-1.13
L21	3	0.57	-	0.05	0.10	0.05
L26	3	0.40	0.20 (1)	-	-	-0.72
L34	3	0.62	0.13	0.03	0.10	0.24
L42	3	0.75	0.37	0.13	0.39	0.65
L45	3	0.29	0.20	0.20	0.60	-1.41
L56	3	0.31	0.05 (1)	0.02	0.10	-1.27
L58	3	0.60	0.17 (1)	0.08	0.10	0.16
L59	3	0.36	0.11	-	-	-0.94
L60	3	0.41	-	0.05	0.06	-0.66
L64	3	0.58	0.17 (1)	0.02	0.10	0.09
L65	3	0.82	0.19	0.18	0.37	0.84
L70	3	0.55	-	0.04	0.08	-0.02
L72	3	0.61	0.37	<=0.04	<=0.08	0.20
L85	3	0.73	0.78	0.10	>0.10	0.59
L88	3	0.50	0.13 (1)	0.10	-	-0.23
			% Incorrect MU	$\mu_{LOD} = 0.08$	$\mu_{LOQ} = 0.18$	$ z_{\mu R} \ge 2.0$
			47%	$\sigma_{LOD} = 0.06$	$\sigma_{LOQ} = 0.17$	0%

(b)

Laboratory Number	Group	Value	Uncertainty	LOD m/m	LOQ m/m	z-score based on µ _R = 0.57
L17	2	0.72	0.28	0.02	0.10	0.56
L20	2	0.57	0.26	0.04	0.08	0.05
L22	2	0.82	0.14	0.10	0.10	0.84
L23	2	0.36	0.02	-	-	-0.94
L24	2	0.71	0.18	0.20	0.40	0.53
L28	2	0.52	0.16	0.05	0.10	-0.15
L29	2	0.70	-	-	-	0.50
L33	2	0.61	0.24	0.04	0.09	0.20
L44	2	2.37	-	-	-	3.15
L48	2	0.47	0.09	0.07	0.15	-0.37
L52	2	1.05	0.07 (1)	0.04	0.20	1.38
L62	2	0.47	0.19 (1)	0.04	0.10	-0.37
L75	2	0.58	0.34	0.02	0.10	0.09
L78	2	0.83	0.15 (2)	0.03	0.30	0.87
L86	2	0.56	0.12	0.30	0.40	0.01
L87	2	0.31	0.10	0.01	0.10	-1.27
			% Incorrect MU	$\mu_{LOD} = 0.07$	$\mu_{LOQ} = 0.17$	% z _{µR} ≥ 2.0
			21%	$\sigma_{LOD} = 0.08$	$\sigma_{LOQ} = 0.11$	6%

(c)

Laboratory Number	Group	Value	Uncertainty	LOD m/m	LOQ m/m	z-score based on $\mu_R = 0.57$
L46	4	0.63	0.15	0.10	0.40	0.27
L55	4	0.43	0.30	0.10	0.10	-0.56
L68	4	0.56	0.28	0.03	0.10	0.01
L76	4	0.68	0.45 (2)	0.10	0.10	0.44
L81	4	0.59	-	0.04	0.08	0.13
L30	5	0.99	0.25	0.01	0.10	1.25
L07	6	0.43	0.13 (1)	0.10	0.10	-0.56
L10	6	0.39	0.08	0.01	0.10	-0.77
L47	6	0.76	0.18	0.05	0.10	0.68
L50	6	0.42	35.60 (3)	0.05	0.10	-0.61
			% Incorrect MU	$\mu_{LOD} = 0.06$	$\mu_{LOQ} = 0.13$	$% z_{\mu R} \ge 2.0$
			33%	$\sigma_{LOD} = 0.04$	$\sigma_{LOQ} = 0.10$	0%

Table A4.2a-c. z-scores for soybean event DP-356043-5 **Level 2 test item** for results reported in m/m %, laboratory category: (a), (b), and (c). -= not reported, (1) Uncertainty (U) and/or coverage factor k was reported in an inconsistent manner, (2) U was reported in an incomplete manner, (3) U seems to be a relative value. Descriptive statistics for LOD and LOQ were estimated using only data for which a value was clearly reported.

(a)

Laboratory Number	Group	Value	Uncertainty	LOD m/m	LOQ m/m	z-score based on µ _R = 1.35
L53	1	1.76	0.55	0.03	0.12	0.63
L01	3	1.51	0.41	-	-	0.30
L03	3	1.37	0.21	-	-	0.09
L04	3	1.41	82.24 (3)	-	-	0.15
L08	3	0.77	30.00 (3)	0.09	-	-1.16
L11	3	0.81	0.32 (1)	-	-	-1.05
L16	3	0.93	0.35 (1)	0.03	0.10	-0.75
L21	3	1.74	-	0.05	0.10	0.61
L26	3	0.90	0.30 (1)	-	-	-0.82
L34	3	1.38	0.24	0.03	0.10	0.11
L42	3	1.03	0.41	0.13	0.39	-0.53
L45	3	0.87	0.40	0.20	0.60	-0.89
L56	3	1.49	0.15 (1)	0.02	0.10	0.27
L58	3	1.15	0.32 (1)	0.08	0.10	-0.29
L59	3	0.91	0.27	-	-	-0.80
L60	3	1.20	-	0.05	0.06	-0.20
L64	3	1.04	0.31 (1)	0.02	0.10	-0.51
L65	3	1.55	0.33	-	-	0.36
L70	3	1.99	-	0.04	0.08	0.90
L72	3	1.26	0.58	<=0.04	<=0.08	-0.09
L85	3	1.99	0.85	0.10	>0.1	0.90
L88	3	1.22	0.31 (1)	0.10	-	-0.16
			% Incorrect MU	$\mu_{LOD} = 0.07$	$\mu_{LOQ} = 0.17$	% z _{µR} ≥ 2.0
			47%	$\sigma_{LOD} = 0.05$	$\sigma_{LOQ} = 0.17$	0%

(b)

Laboratory Number	Group	Value	Uncertainty	LOD m/m	LOQ m/m	z-score based on µ _R = 1.35
L17	2	1.29	0.34	0.02	0.10	-0.04
L20	2	1.27	0.45	0.04	0.08	-0.07
L22	2	1.78	0.17	0.10	0.10	0.66
L23	2	1.18	0.52	-	-	-0.23
L24	2	1.37	0.07	0.20	0.40	0.09
L28	2	1.41	0.21	0.05	0.10	0.15
L29	2	1.30	-	-	-	-0.02
L33	2	1.46	0.27	0.04	0.09	0.23
L44	2	3.32	-	-	-	2.02
L48	2	1.06	0.20	0.07	0.15	-0.46
L52	2	2.00	0.20 (1)	0.04	0.20	0.91
L62	2	1.38	0.57 (1)	0.04	0.10	0.11
L75	2	0.82	0.20	0.02	0.10	-1.02
L78	2	1.91	0.34 (2)	0.03	0.30	0.81
L86	2	1.70	0.22	0.30	0.40	0.56
L87	2	1.25	0.42	0.01	0.10	-0.11
			% Incorrect MU	$\mu_{LOD} = 0.07$	$\mu_{LOQ} = 0.17$	% z _{µR} ≥ 2.0
			21%	$\sigma_{LOD} = 0.08$	$\sigma_{LOQ} = 0.12$	6%

(c)

Laboratory Number	Group	Value	Uncertainty	LOD m/m	LOQ m/m	z-score based on $\mu_R = 1.35$
L46	4	1.14	0.25	0.10	0.40	-0.31
L55	4	1.82	1.53	0.10	0.10	0.71
L68	4	1.00	0.50	0.03	0.10	-0.59
L76	4	1.65	1.07 (2)	0.10	0.10	0.50
L81	4	1.76	-	0.04	0.08	0.64
L30	5	2.07	0.52	0.01	0.10	0.99
L07	6	1.09	0.33 (1)	0.10	0.10	-0.40
L10	6	0.84	0.10	0.01	0.10	-0.97
L47	6	1.13	0.25	0.05	0.10	-0.33
L50	6	1.22	35.60 (3)	0.05	0.10	-0.16
			% Incorrect MU	$\mu_{LOD} = 0.06$	$\mu_{LOQ} = 0.13$	% z _{µR} ≥ 2.0
			33%	$\sigma_{LOD} = 0.04$	$\sigma_{LOQ} = 0.10$	0%

Table A4.3. z-scores for soybean event DP-356043-5 **Level 1 test item** for results reported in cp/cp %, all laboratory categories. * z-score not calculated, - = not reported, (1) Uncertainty (U) and/or coverage factor k was reported in an inconsistent manner, (2) U was reported in an incomplete manner, (3) U seems to be a relative value.

Laboratory Number	Group	Value	Uncertainty	LOD cp/cp LOQ cp/cp		z-score based on µ _R = 0.64
L27	2	0.89	4.00 (2) (3)	0.05	0.10	<u>μκ = 0.54</u> 0.77
L31	2	8.49	4.00 (2) (3) 3.27	0.03	0.10 0.43	5.6 7
L37	2	0.68	0.23 (1)	0.04	0.08	0.18
L71	2	0.63	0.09	0.30	0.60	0.02
L74	2	< 0.90	-	-	0.42	*
L18	3	< 0.70	-	0.20	0.80	*
L19	3	0.38	0.14 (1)	0.05	0.10	-1.08
L61	3	0.50	0.18	-	0.10	-0.48
L02	6	0.61	0.05	-	-	-0.05
L67	6	0.50	1.05	-	0.04	-0.48
L77	6	> 0.10	-	0.10	-	*
			% Incorrect MI	$\mu_{LOD} = 0.13$	$\mu_{LOQ} = 0.30$	$% z_{\mu R} \ge 2.0$
			38%	$\sigma_{LOD} = 0.10$	$\sigma_{LOQ} = 0.28$	13%

Table A4.4. z-scores for soybean event DP-356043-5 **Level 2 test item** for results reported in cp/cp %, all laboratory categories. * z-score not calculated, - = not reported, (1) Uncertainty (U) and/or coverage factor k was reported in an inconsistent manner, (2) U was reported in an incomplete manner, (3) U seems to be a relative value.

Laboratory Number	Group	Value	Uncertainty	LOD cp/cp	LOQ cp/cp	z-score based on $\mu_R = 1.37$
L27	2	1.45	7.00 (2) (3)	0.05	0.10	0.13
L31	2	8.46	1.56	0.18	0.43	3.96
L37	2	1.30	0.39 (1)	0.04	0.08	-0.11
L71	2	1.58	0.03	0.30	0.60	0.32
L74	2	> 0.9	-	-	0.42	*
L18	3	0.79	0.09	0.20	0.80	-1.19
L19	3	1.17	0.19 (1)	0.05	0.10	-0.34
L61	3	1.46	0.93	-	0.10	0.14
L02	6	1.23	0.12	-	-	-0.23
L67	6	1.42	1.05	-	0.04	0.08
L73	6	< 0.08	-	0.04	0.09	*
L77	6	> 0.10	-	0.10	-	*
			% Incorrect MU	$\mu_{LOD} = 0.12$	$\mu_{LOQ} = 0.28$	$ z_{\mu R} \ge 2.0$
			33%	$\sigma_{LOD} = 0.10$	$\sigma_{LOQ} = 0.27$	11%

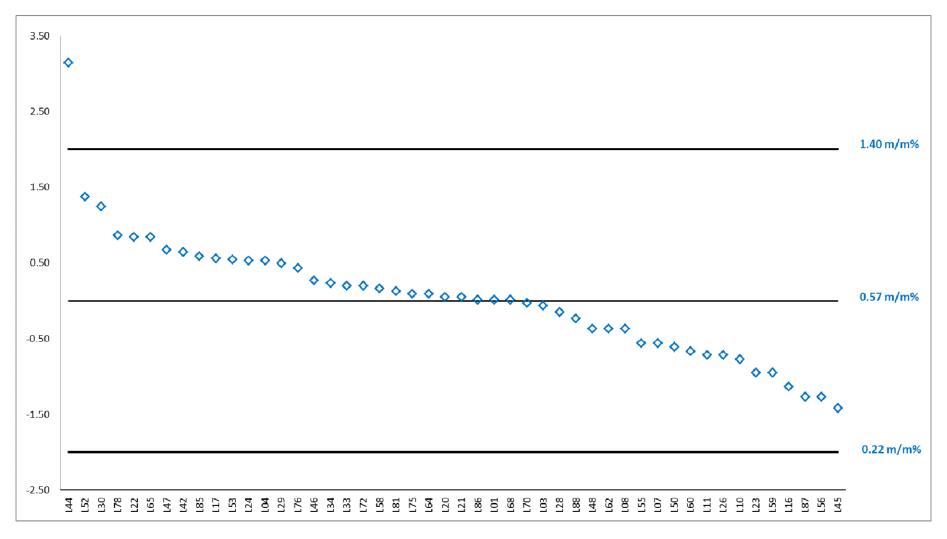


Figure A4.1. z-scores for soybean event DP-356043-5 Level 1 test item on the basis of a robust mean of 0.57 m/m % (\$).

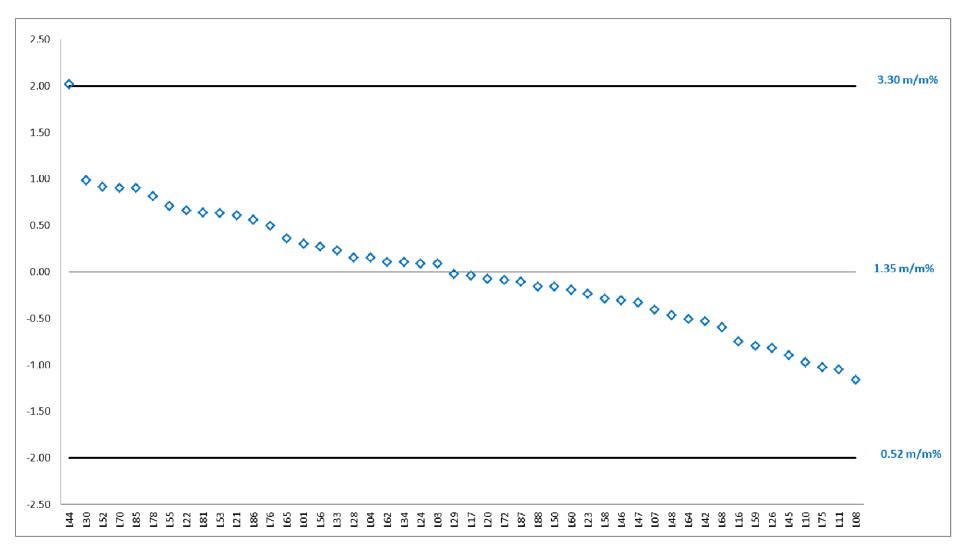


Figure A4.2. z-scores for soybean event DP-356043-5 Level 2 test item on the basis of a robust mean of 1.35 m/m % (0).

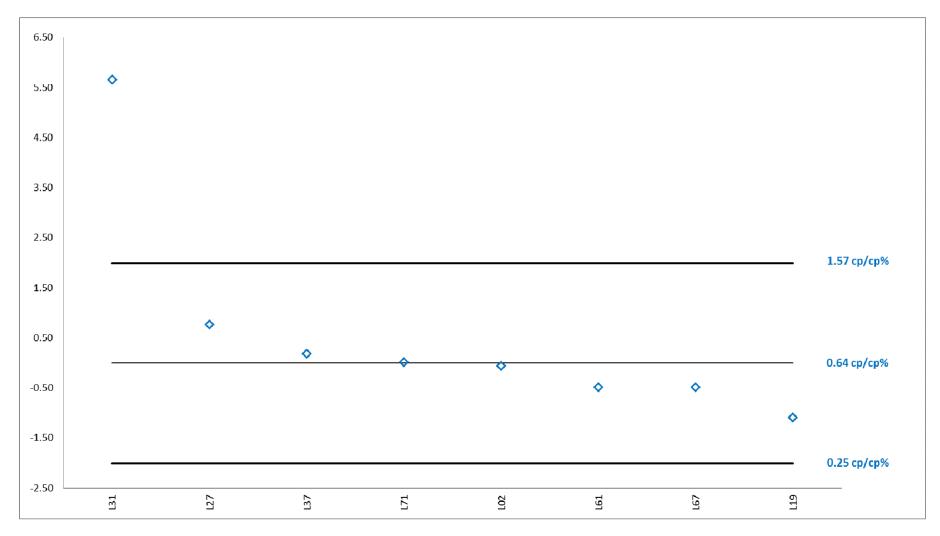


Figure A4.3. z-scores for soybean event DP-356043-5 Level 1 test item on the basis of a robust mean of 0.64 cp/cp % (◊).

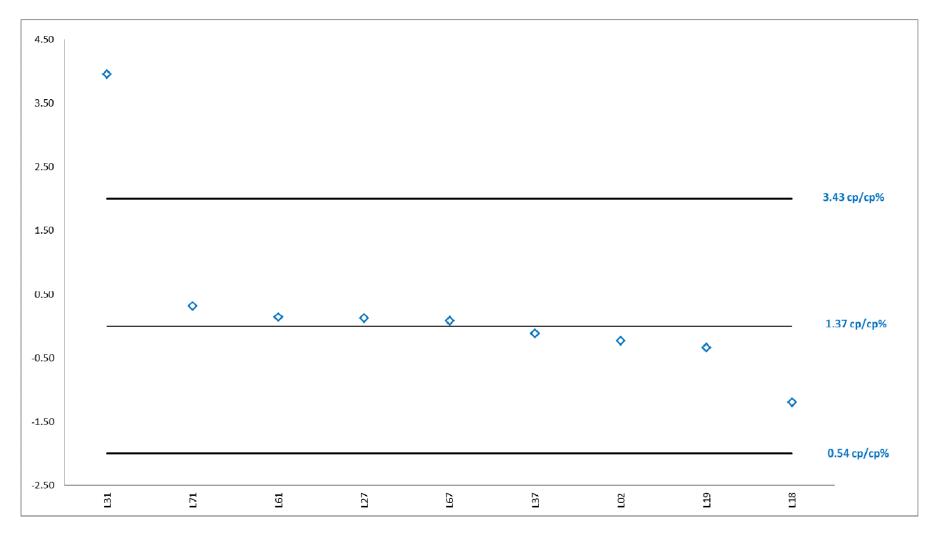


Figure A4.4. z-scores for soybean event DP-356043-5 Level 2 test item on the basis of a robust mean of 1.37 cp/cp % (◊).

Annex 5: Questionnaire data

Q1.1. Rice noodles Sample L1:	No. of laboratories per species			
species identification	Maize	Oilseed Rape	Rice	Soybean
Present	8	1	80	85
Absent	77	73	0	0
Not Tested	0	11	5	0

Q1.2. Rice noodles Sample L2: species identification	No. of laboratories per species			
	Maize	Oilseed Rape	Rice	Soybean
Present	11	4	78	81
Absent	74	70	2	4
Not Tested	0	11	5	0

		No.	of laboratorie	es per Sampl	е	
Q2. GM event(s) identification	GM E	ent 1	GM E	vent 2	GM E	vent 3
identification	Sample 1	Sample 2	Sample 1	Sample 2	Sample 1	Sample 2
DP-356043-5	62	63	0	0	0	0
p35s	5	5	2	2	0	0
Soybean Line GTS 40-3-2	2	2	0	0	0	0
MON810	1	1	0	0	0	0
NOS	0	0	1	1	0	0

3. Number of replicate DNA extractions from test materials:	No. of laboratories
a) 1	1
b) 2	59
c) 3	9
d) 4	10
e) Other	6
Other of which:	
5	1
6	4
8	1

4. DNA extraction method:	No. of laboratories	
a) ISO/CEN	31	
b) EU-RL	3	
c) National reference method	6	
d) International literature	2	
e) In-house developed and optimised	8	

f) Commercial kit	29
g) Other	6
Other of which	Answers referred to used kits, see Q6

4.3. Is the DNA extraction method used under ISO/IEC 17025 accreditation?	No. of laboratories
a) Yes	71
b) No	14

5. Sample intake (in g) for the DNA extraction?	No. of laboratories
a) < 0.1	3
b) 0.1 - 0.2	44
c) > 0.2	30
d) Other	8
Other of which:	
0.2	1
1	4
2	3

6. DNA extraction method/kit used?	No. of laboratories
a) CTAB	38
b) CTAB-derived	13
c) Biotecon	4
d) GeneScan GENESpin	3
e) Guanidine HC1 with proteinase K	2
f) Macherey-Nagel Nucleospin	11
g) Promega Wizard	4
h) Qiagen Dneasy plant mini kit	4
i) TEPNEL kit	1
j) In-house developed and optimised	1
k) Other	4
Other of which:	
PowerPlant Pro DNA Isolation Kit, MoBio Laboratories, Inc.	1
SureFood PREP Animal X-Kit by r-biofarm	1
Generon Ion Force Fast	1
Modified SDS DNA extraction method	1

7. How was the clean-up of the DNA performed?	No. of laboratories	
a) No DNA clean-up	41	
b) Ethanol precipitation	16	
c) Amersham MicroSpin S300	0	
d) Promega Wizard DNA clean-up resin	5	

e) Qiagen QIAQuick	11	
f) Qiagen Genomic-Tip 20/G	1	
g) Silica	7	
h) Other	4	
Other of which:		
Maxwell 16 Nucleic Acid Extraction Kit (Promega)	1	
As part of the CTAB method, the DNA is purified by isopropanol precipitation and washing with ethanol.	1	
Invisorb DNA Cleanup	1	
DNA Extractor Cleaning Columns Kit (eurofins)	1	

8. How have you quantified the genomic DNA extracted?	No. of laboratories
a) Gel	2
b) UV spectrophotometer	64
c) Fluorometer	11
d) Other	3
e) Not applicable (i.e. DNA was not quantified)	5
Other of which:	
Estimation is based on qPCR results.	1
Nanodrop	1
QPCR	1

9. DNA Dilution buffer	No. of laboratories
a) TE (10 mM Tris-HC1, 1 mM EDTA)	18
b) TE 0.1X (10 mM Tris-HC1, 0.1 mM EDTA)	13
c) TE low (1 mM Tris, 0.01 mM EDTA)	2
d) Water	42
e) Other	10
Other of which	
Buffer EB ,from Qiagen mericon food kit	1
0.5 X TE	1
TE 0,2x	1
TE (10 mM TrisHCl; 0,2 mM EDTA)	1
Elution Buffer Part# A 828D (Maxwell 16 FFS Nucleic Acid Extraction Kit / Promega)	1
0.2x TE (2 mM Tris-HCl, 0.2 mM EDTA)	1
PE (5 mM Tris HCl, pH 8.5); from Nucleospin plant II kit	1
BE buffer from QIAquick kit	1
Buffer EB	1
No dilution applied	1

10. Screening methods used for GMO detection	No. of laboratories
a) Reference method from EU-RL GMFF GMOMETHODS database	31
b) National reference method	17
c) ISO/CEN	27
d) In-house developed and optimised	8
e) National reference method	3
f) Pre-spotted plate	6
g) Other	22
Other of which:	
CoSYPS methods - will included in the EU-RL GMFF database	1
Commercial kit	2
international literature	3
Screening table; BVL	1
SybrGreen Q PCR (CoSYPS method")	1
Pentaplex Screening PCR, GMOseek project	1
not used	1
Eur Food Res Technol (2008) 226(5):1221-1228; J. Agric. Food Chem (2009) 57(19):8913-8920; J AOAL Int (2002) 85(3): 646-653	1
§ 64 00.00-122; 00.00-124; 00.00-125; § 64 L 15.06-03; Forschungsprogramm "Ernaehrung/Nahrungsmittelsicherheit" der Landesstiftung Baden-Württemberg (2007) Molekularbiologische Verfahren zum Nachweis von nicht-zugelassenen gentechnisch veränderten Pflanzen. Abschlussbericht. Projekt P-LS-E2/11.	1
Eurofins GMOScreen 35S/NOS/FMV IPC	1
Eurofins GeneScan	1
Real time PCR, see below	1
Reiting R., Grohmann L., Dietrich Maede D. Journal of Consumer Protection and Food Safety Vol.5 N°2, 185-188, 2010 "Testing cascade for the detection of genetically modified rice by real-time PCR in food and its application for detection of an unauthorized rice line similar to KeFeng6	1
Debode et al. (2013) and Kuribara et al. (2002); Bayer Cropscience (2006)	1

10.2 Is the PCR screening method used under ISO/IEC 17025 accreditation?	No. of laboratories
a) Yes	70
b) No	15

11. Principle of PCR detection used for screening?	No. of laboratories
a) Gel	7
b) SYBR [®] Green	4
c) Taqman probe	69
d) Other	5
Other of which:	
Gel, Taqman probe	1
Gel and SYBR Green and Taqman	1
not used	1
CryIAb/CryIAc-SYBR-Green; p35S/tNOS +bar+PLD+cruA - Taqman probe,lectin +ivr-gel	1
GEL, TAQMAN PROBE	1

12. Screening method used for GM detection:	No. of laboratories
a) Multiplex PCR	23
b) Singleplex PCR	62

13. Elements / targets used for screening (P = promoter, T = terminator):	No. of laboratories
a) P35S	76
b) T-nos	74
c) Other	58
Other of which:	
pFMV	5
bar	2
PAT, BAR	1
CTP2-CP4-EPSPS, bar, 35S-pat, CryIAb/Ac (SG), CaMV, 305423, 356043, CV127, MON87701, MON87708,	1
pFMV and P35S-BAR	1
NPTII, Bar	1
pat, cp4-epsps, adh, cruciferine, PLD, Lectine	1
Cry1Ab, pat/bar, EPSPS	1
pat, rActin intron bar, nptII, cry1Ac, cry1Ab/Ac	1
CP4EPSPS	1
Endogenous Genes	1
bar, FMV, ctp2-cp4-epsps, pat	1
pFMV, npt II, pat, bar,	1
CP4-EPSPS, NPTII, PAT, CP4CTP-EPEPS	1
CTP2	1
pat, bar, CTP2-CP4EPSPS, cry1ab/ac	1
Cry A(c)-T-Nos construct	1
Screening table; BVL	1
CPT2-CP4 EPSPS, Cry1Ab/Ac P-ubi-cry	1
FU DI CMEE C	40.450

event-specific	1
Duplex PCR: p35S/T-nos, bar/p35S-pat, Singleplex PCR: cry1Ab/Ac	1
35Spat;CTP2-CP4EPSPS;cryIA(b)	1
pat, bar, ctp2-cp4-epsps	1
P-FMV, bar, pat, nptII, Cry1Ab, Cry1Ac	1
not used	1
CTP2-CP4-EPSPS; bar; pat	1
p35S/tNOS +Cry IAb/CryIAc + bar	1
CT4EPSPS, bar, 35S-pat	1
P-FMV, Cry1Ab/Ac	1
pat, bar, cryIAb/IAc	1
T35s, CTP2-CP4	1
Multiplex: P35s / Tnos; Singleplex: bar, pat, Pnos, cryIAb/Ac	1
CP4 EPSPS	1
P34S-FMV	1
cryAb/Ac SyberGreen	1
P35S-PAT,	1
Cry1Ab/Ac	1
ctp2-cp4epsps; BAR; PAT, 35S-PAT	1
Pat, Bar, CP4 EPSPS	1
see 10.3.	1
NPTii, CP4EPSPS, CTP-CP4EPSPS, PAT	1
event specific	1
Cry1Ab - SYBR Green	1
EU-RL pre-spotted plate: Querci et al., 2009	1
T-35S, cry1Ac, epsps (Based on the screening results, both the samples L1 and L2 were found negative to these targets).	1
CP4 EPSPS, 35S-pat;cry 1Ab; pubi-cry	1
cry1Ab/Ac;	1
PAT; NPTII; CTPCP4-EPSPS; CP4-EPSPS	1
NPTII, RRS	1
cryIA(b and c), bar	1
Trait Specific primers and probe	1

14. Construct-specific methods used for GMO detection	No. of laboratories
a) Reference method from EU-RL GMFF GMOMETHODS database	28
b) National reference method	15
c) ISO/CEN	14
d) In-house developed and optimised	3

e) International literature	9
f) Other	30
Other of which:	
Not applicable	1
None/Not Used/ Not Tested	12
Pre-Spotted Plate	1
see screening	1
CTP2-CP4EPSPS	1
Event specific	1
Screening table; BVL	1
not applicable	1
Methods Waiblinger (Germany) - Screening table	1
CTP2-CP4EPSPS + Bar	1
35S/BAR	1
Eurofins GeneScan GMOQuant Roundup Ready soy	1

14.3. Real-time PCR quantification method used under ISO/IEC 17025 accreditation?	No. of laboratories
a) Yes	49
b) No	22
Missing	14

15. Principle of PCR used for construct-specific detection	No. of laboratories
a) Gel	7
b) SYBR ® Green	4
c) Taqman probe	55
d) Other	4
Missing	20
Other of which:	
Not used	4

16. Construct-specific method:	No. of laboratories
a) Multiplex PCR	8
b) Singleplex PCR	76
Missing	1

17. Please specify targets used for construct specific detection (P = promoter, T = terminator)	No. of laboratories
Not done/Not used	8
p35S	1
pSAMS : gm-hra gene	1
CTP2-CP4EPSPS	2

CTP4-CP4EPSPS	1
ctp4-cp4epsps and ctp2-cp4epsps	1
RR	1
pubi-cry/35S-hpt/cpti-Nos	1
RRS, LLRICE 601, LL62	1
P35S-BAR	1
pUbi-cry, 35S-hpt, T51-1 (Bt63), 35S-pat, CTP2-CP4EPSPS	1
Cry A(c)-T-Nos construct	1
CTP2-EPSPS; 35S-pat	1
Р, Т	1
§64 LFGB L 00.00-125 (CTP2-CP4-EPSPS), L 15.06-1 (cry1Ac-T-nos), §28b GenTG G 30.40-1 (Duplex PCR: p35S-pat with bar)	1
ctp2-cp4epsps;P35S-pat	1
P35S and T-NOS	1
CP4-EPSP; CTP-CP4EPSPS	1
CTP2-CP4EPSPS; p35s-pat;	1
35S-pat, CTP2-cp4epsps	1
Bt63	1
CTP2-CP4EPSPS + Bar	1
P35s, TNOS, T35s, CTP2-CP4	1
CTP2-CP4-EPSPS, p35S-pat, pubi-cry, Cry1Ab/1Ac	1
CTP2-CP4EPSPS, 35s-pat, Pnos-nptII	1
P	1
cryI(Ac)/T-nos	1
P35S, TNOS, Cry IAb/Ac, pat,bar	1
Event specific	1
CTP2-CP4-EPSPS; cryIA(c)::Nos; pUbi-cry	1
pat, bar, cry1Ab/Ac	1
P35S/pat; P35S/bar; CTP2-CP4EPSPS; pubi-cry; 35S/htp; cpti-nos; cry-Tnos (KeFeng6); cryIA(c)/nosT (Bt63);	1
Р, Т	1
CP4EPSPS, CTP-CP4EPSPS, NPTII, PAT	1
35S PRomoter, Nos 3' Terminator, nptII,RRS	1

18. Event-specific methods used for GMO detection	No. of laboratories
a) Reference method from EU-RL GMFF GMOMETHODS database	70
b) National reference method	5
c) ISO/CEN	6
d) In-house developed and optimised	1
e) International literature	1
f) Other	5

Missing	4
Other of which:	
Event-specific Pre-Spotted Plates	1
Event-specific method for Rice: LLRice601 (Bayer CropScience),	1
GMOIdent Event MON810 Corn GeneScan	1
no construct-specific methods used	1
AllSoy02-Kit (Microsynth, CH): GTS40-3-2; MON89788; A2704-12; A5547-127	1

18.3. Is the PCR event-specific method used under ISO/IEC 17025 accreditation?	No. of laboratories
a) Yes	54
b) No	23
Missing	8

19. Principle of PCR used for event-specific detection	No. of laboratories	
a) Gel	6	
b) SYBR ® Green	1	
c) Taqman probe	74	
d) Other	0	
Missing	5	

20. Event-specific method	No. of laboratories	
a) Multiplex PCR	5	
b) Singleplex PCR	58	
Missing	22	

21. Digital PCR quantification method(s)	No. of laboratories	
a) In-house developed and optimised	5	
b) International literature	0	
c) National reference method(s)	5	
d) Other	72	
Missing	4	
Other of which:		
Not used/Not Tested/not applied/etc	28	

21.3. Is the digital PCR quantification method used under ISO/IEC 17025 accreditation?	No. of laboratories
a) Yes	5
b) No	19
Missing	61

22. Real-time PCR instrument:	No. of laboratories
a) ABI 7000	1
b) ABI 7300	10
c) ABI 7500	34
d) ABI 7700	1
e) ABI 7900 HT	18
f) ABI StepOne & StepOne Plus real-time PCR system	3
g) BioRad icycler	2
h) Corbett Rotor-Gene 6000	2
i) Roche LightCycler 480	3
j) Roche Light Cycler 2.0	1
k) Stratagene Mx 3000/Mx 3005	7
I) Stratagene Mx4000	0
m) other	14
Other of which:	
ViiA7	2
Roche Light Cyler 1.2	1
Rotor-Gene 3000 (Corbett Research), iQ5 (BioRad)	1
not tested	1
BioRad CFX96	6
BIORAD CFX	2
Qiagen Rotor-Gene Q	2

23. Real-time PCR Master Mix:	No. of laboratories
a) ABI TaqMan [®] Universal PCR master mix	45
b) ABI TaqMan [®] Universal PCR master mix, no AmpErase [®] UNG	7
c) ABI TaqMan® Fast Universal PCR master mix	2
d) ABI TaqMan [®] Gold with Buffer A	1
e) Eurogentec: qPCR MasterMix	4
f) Eurogentec MESA GREEN qPCR MasterMix Plus for SYBR® Assay	0
g) Eurogentec qPCR MasterMix for SYBR® Green	0
h) Sigma JumpstartTM Taq ReadyMixTM	2
i) Qiagen: QuantiTect SYBR® Green PCR Kit	0
j) Qiagen: QuantiTect Probe PCR Kit	4
k) Roche: FastStart TaqMan® Probe Master (Rox)	2
l) Roche: FastStart Universal Probe Master (Rox)	0
m) Diagenode: Universal Mastermix	2
n) Fermentas: Maxima [™] Probe/ROX qPCR Master Mix	2
o) Fermentas: Maxima [™] SYBR Green/ROX qPCR Master Mix	1
p) Ampliqon: RealQ PCR 2 x Master Mix	0

q) Takara: SYBR [®] Premix Ex Taq [™]	0
r) Takara: Premix Ex Taq [™]	0
s) Other	25
Missing	1
Other of which:	
5x Hot Fire PoI probe qPCR master mix (Bioconnect)	1
ABI SybrGreen Master Mix	1
additionally: QuantiTect Multiplex NoROX PCR Kit (Qiagen)	1
AmpliTaq gold with Buffer II	1
Bioline SensiFAST Probe Lo-ROX Mix	1
BioRad iQ Sybr Green Supermix	1
BioRad Supermix or Powermix	
Brilliant III QPCR & QRT-PCR Master Mix, Agilent tech.	1
comercial kit, MasterMix eventMON810 provided by GMOQuant EventMON810 Corn GeneScan	1
Eurofins GeneScan	1
Eurofins Reaction mix, PowerSYBR® Green PCR Master Mix	1
GENETICBIO HOTSTART TAQ DNA POLYMERASE	1
GoTaqProbe qPCR MasterMix by Promega	1
KAPA Prope Fast Universal	1
not tested	1
PROMEGA GoTaq qPCR MasterMix	1
Qiagen quantiFast Multiplex PCR	1
Qiagen: QuantiTect Multiplex NoROX Master Mix	1
Qiagen: QuantiTect Multiplex RT-PCR NoROX Master Mix	1
Quanta Biosciences Perfecta qPCR Master Mix	1
Roche Applied Science. Light Cyler GMO Soya Quantification Kit	1
Roche fast start master hybridization probe	1
Roche LightCycler 480 Probes Master (Cat.No. 04 887 301 001)	1
Termo Scientific Maxima Probe /Rox qPCR MAsterMix (2x)	1
UMTS	1

23.2. Number of reagents (<i>i.e.</i> DNA, primers, probe, water,) involved?	No. of laboratories
a) 5	42
b) 6	31
c) 7	6
d) 8	1
e) other	5
Missing	1
Other of which:	

0	1
3	1
15	1
17	1
38	1

Q24.1 Sample intake (in ng) per real-time PCR	No. of laboratories
a) 0-50	15
b) 50-100	24
c) 100-200	32
d) > 200	4
e) DNA amount not quantified	7

Q24.2. Sample intake (in ng) per real-time PCR	No. of laboratories
a) 0-50	4
b) 50-100	2
c) 100-200	7
d) > 200	2
e) DNA amount not quantified	2

Q24.3. Sample intake (in ng) per real-time PCR	No. of laboratories
a) 0-50	2
b) 50-100	1
c) 100-200	4
d) > 200	1
e) DNA amount not quantified	1

Q24.4. Sample intake (in ng) per real-time PCR	No. of laboratories
a) 0-50	2
b) 50-100	1
c) 100-200	3
d) > 200	0
e) DNA amount not quantified	1

Q24.5. Sample intake (in ng) per real-time PCR	No. of laboratories
a) 0-50	1
b) 50-100	1
c) 100-200	3
d) > 200	0
e) DNA amount not quantified	1

25. Real-time PCR quantification method(s) used?	No. of laboratories

a) DNA copy number standard curve using a dilution series 24

b) Mass/mass standard curve using a dilution series	43
c) Delta Ct method	9
d) Absolute quantification (end-point digital PCR)	1
e) Other	7
Missing	2
Other of which	
No quantification performed	7

Q26. Real-time PCR	No. of laboratories per GM event		
quantification method(s): slope(s) endogenous gene	GM event 1	GM event 2	GM event 3
-4.1 ≤ slope < -3.6	2	0	0
$-3.6 \le \text{slope} \le -3.1$	55	3	1
-3.1 < slope < -2.6	3	0	0
Other	1	0	0
Not Applicable	1	0	0

Q27. Real-time PCR quantification method(s): – slope(s) GM trait gene	No. of laboratories per GM event		
	GM event 1	GM event 2	GM event 3
-4.1 ≤ slope < -3.6	3	0	0
$-3.6 \le \text{slope} \le -3.1$	55	3	1
-3.1 < slope < -2.6	2	1	0
Other	1	0	0
Not Applicable	1	0	0

Q28. Real-time PCR quantification method(s): R ² coefficient(s) endogenous gene	No. of laboratories per GM event		
	GM event 1	GM event 2	GM event 3
$0.97 \le R^2 < 0.98$	2	0	0
$0.98 \le R^2 \le 0.99$	9	1	0
$0.99 < R^2 < 1.00$	51	2	1
Other	1	0	0
Not Applicable	0	0	0

Q29. Real-time PCR quantification method(s):	No. of laboratories per GM event			
R ² coefficient(s) GM trait gene	GM event 1	GM event 2	GM event 3	
$0.97 \le R^2 < 0.98$	5	0	0	
$0.98 \le R^2 \le 0.99$	14	0	0	
$0.99 < R^2 < 1.00$	44	3	1	
Other	1	0	0	
Not Applicable	0	0	0	

Q30. Real-time PCR quantification method(s):	No. of	1 event	
endogenous target DNA sequence(s)	GM event 1	GM event 2	GM event 3
Lectin	14	1	0
Lectin (74 bp)	29	0	0
Lectin (80 bp)	1	0	0
Lectin (81 bp)	5	0	0
Lectiin (102 bp)	2	0	0
Lectin (105 bp)	1	0	0
Lectin (118 bp)	3	0	0
SSIIb-3	1	0	0
CruA	1	0	0
TM013	1	0	0
nptII	1	1	0
HMG	1	0	0
HMG (79 bp)	1	0	0
invertase	1	1	0
sps (81 bp)	1	0	0
p35S	1	1	0

Q31. Real-time PCR quantification method(s):	No. of laboratories per GM event				
GM trait target DNA sequence(s)	GM event 1	GM event 2	GM event 3		
DP356043	15	0	0		
DP356043 (74 bp)	1	0	0		
DP356043 (83 bp)	1	0	0		
DP356043 (99 bp)	34	1	0		
p35s	1	0	0		
p35S, 82bp	2	1	0		
specific for	1	0	0		
RUR GTS 40-3-2 84	1	0	0		
RR (83 bp)	1	0	0		
MON810	1	0	0		

Q32. Which reference material(s) was (were) used for calibration?	No. of laboratories
ERM	5
ERM-AD425	1
ERM-BF425 series	9
ERM-BF425a	1
ERM-BF425b	1

ERM-BF425c	4
ERM-BF425d	38
ERM-BF410 series	1
ERM-BF410dk	1
ERM-BF410gk	2
Eurofins GeneScan	2
10% GTS 40-3-2	1
Single target	1
426d	1
biosmart exStGVO-	1
ERM-BF series	1
plasmid	1
EURL Plasmids	1
AOCS 034-B	1
EURL Stds from	1
AOCS 0906-B	1
Commercially	1

Q33. Which reference material(s) was (were) used for quality control, <i>e.g.</i> bias control??	No. of laboratories
0.1% GTS 40-3-2	1
0.5% GTS 40-3-2	1
1% GTS 40-3-2	1
2% GTS 40-3-2	1
5% GTS 40-3-2	1
AOCS 0210-A	1
AOCS 0306-I	1
AOCS 0306-I2	1
AOCS 0306I4 (LL62)	1
AOCS 0311-A	1
AOCS 034-B	1
AOCS 0707B	1
AOCS 0707-B4	1
AOCS 0707C	1
AOCS 0707-C3	1
AOCS 0809-A	1
AOCS 0906B	3
AOCS 0911-C	1
AOCS 208 series	1
AOCS 306 SERIES	1
AOCS 707B	1

AOCS 707-C AND	1
AOCS 804 series	1
Commercially	1
Control sample	3
CRL-40-3-2	1
CRL-A2704	1
CRL-DP356043	1
CRL-FG72	1
CRL-MON89788	1
CRM maize Bt 11;	1
CRM soya GTS40-3-	1
DNA potato 100%	1
356043 soya ERM-	2
ERM-BF series	1
ERM-BF410 series	3
ERM-BF410bk	2
ERM-BF410dk	1
ERM-BF410gk	2
ERM-BF411	1
ERM-BF412	1
ERM-BF412d	1
BF 413d	1
ERM-BF 414 series	1
ERM-BF415	1
ERM-BF425 serie	1
ERM AD425	1
ERM BF 425	3
ERM-BF425a	3
ERM BF 425b	16
ERM BF 425c	26
ERM BF 425d	11
426b	2
426c	2
EURL Plasmids	1
EURL Stds from	1
EURL-GMFF	2
FLUKA IRMM	1
GeneScan BTRICE PC	1
GT73/RT73 Canola	1
LL RICE 62	1
Maize NK603 ERM-	1

Maize powder 1%	1	
none	1	
PENGL-00-	1	
pENGL-00-04/07-	1	
Rice Leaf DNA 100%	1	
Roundup Ready	1	
Single target	1	
Soybean powder 1%	1	
Sugar beet powder	1	

Q34a. Practical LOD and LOQ (in %) of the GM content	No. of laboratories - GM event 1				
determination in mass/mass or DNA copy number ratio?	LOD (m/m)	LOQ (m/m)	LOD (cp/cp)	LOQ (cp/cp)	
0.01	4		1	-	
0.02	4				
0.025	4				
0.039				1	
0.04	8		1		
0.05	5		1		
0.06		1		1	
0.07	1				
0.08	1	4			
0.09	1	2		1	
0.10	7	21	2	4	
0.15		1			
0.18	1		1		
0.20	2	1	1		
0.30	1	1	1		
0.33	1				
0.37		1			
0.39		1			
0.40		1			
0.42				1	
0.43				1	
0.60		1		1	
0.80				1	
<=0.04	1				
<=0.08		1			
>0.10		1			
L1 0.027; L2 0.030	1				
L1 0.106; L2 0.121		1			

Not reported	43	47	77	74
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Q34b. Practical LOD and LOQ (in %) of	No. of laboratories - GM event 2			
the GM content determination in mass/mass or DNA copy number ratio?	LOD (m/m)	LOQ (m/m)	LOD (cp/cp)	LOQ (cp/cp)
0.005	1			•
0.20	1			
0.29			1	
0.58				1
0.60		1		
Not reported	83	84	84	84

35.1. Does the uncertainty correspond to a repeatability standard deviation?	No. of laboratories
a) Yes	40
b) No	10
c) Not applicable	14
Not reported	21

35.2. Does the uncertainty correspond to a within-laboratory reproducibility standard deviation?	No. of laboratories
a) Yes	25
b) No	24
c) Not applicable	12
Not reported	24

35.3. Does the uncertainty include a contribution from the DNA extraction step?	No. of laboratories
a) Yes	31
b) No	19
c) Not applicable	11
Not reported	24

35.5. Did you report an expanded uncertainty including a coverage factor?	No. of laboratories
a) Yes	48
b) No	4
c) Not applicable	12
Not reported	21

35.6. If applicable, please specify the coverage factor used (k = 1 for a 66.67% confidence level, k = 2 for a 95% confidence level, k = 3 for a 99% confidence level):	No. of laboratories
a) k = 1	1
b) k = 2	46
c) k = 3	1
d) Other	5
Not reported	32
Other of which:	
The coverage factor depend on the number of measurements (n=10 factor 2.262 and n=26 factor 2.060) for P=95% and f=n-1	1
2.78	1
Coverage factor dependent upon degrees of freedom and vary from 2.26 (9 d.f.) to 2.20 (11 d.f.)	1
k = 2.57 for sample L2, $k = 2.78$ for sample L1	1

Annex 6: Invitation letter





Ispra, 28 March 2014 JRC.DG.I.3/MBG/JK/mm/lv

NOTE FOR THE ATTENTION OF

- I. All National Reference Laboratories nominated under COMMISSION REGULATION (EC) No 882/2004
- II. All National Reference Laboratories nominated under COMMISSION REGULATION (EC) No 1981/2006
- III. All members of the European Network of GMO Laboratories
- IV. Official control laboratories
- V. Interested parties from third countries

Subject: Invitation to participate in the comparative test ILC-EURL-GMFF-CT-02/13

Pursuing Regulation (EC) No 882/2004 of the European Parliament and of the Council of 29 April 2004, the European Union Reference Laboratory for GM Food and Feed (EU-RL GMFF) has the obligation to organise comparative testing rounds and to ensure an appropriate follow-up of the results obtained.

Hereby, I would like to invite you to participate in the eighth round of comparative testing ILC-EURL-GMFF-CT-02/13. This round of comparative testing will include two test materials of rice noodles. Participants will have to:

- Perform species identification: maize, soybean, oilseed rape and rice
- Identify and quantify the GM event(s) detected.

Your participation is free of charge. Participants in the comparative testing rounds need to dispose over equipment for qualitative and quantitative Polymerase Chain Reaction (PCR).

I would like to remind you that participation in comparative testing is mandatory for all National Reference Laboratories nominated under Regulation (EC) No 882/2004. The participation of National Reference Laboratories nominated under Regulation (EC) No 1981/2006 is not mandatory though highly recommended. The laboratory codes assigned to each participant in this comparative testing round are confidential. However, the EU-RL GMFF will disclose details of the National Reference Laboratories that have been appointed under Regulation (EC) No 882/2004 to DG SANCO for the purpose of an assessment of their performance.

ISO 9001:2008 certified by



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Registration for the eighth round of comparative testing and submission of results will be handled by the EU-RL GMFF. Please register electronically using the following link:

https://web.jrc.ec.europa.eu/ilcRegistrationWeb/registration/registration.do?selComparison=1261

Please be aware that each laboratory can only register once for this comparative testing round. Hence it will no longer be possible to submit your results in both measurement units (i.e. mass/mass % and cp/cp %) for the same set of test items.

The deadline for registration is <u>4 April 2014</u>. Samples should be shipped during the week of <u>14-17 April 2014</u>. The deadline for submission of results is <u>30 May 2014</u>.

Please contact the functional mailbox mbg-comparative-testing@jrc.ec.europa.eu for all issues related to this comparative testing round, e.g. difficulties with your on-line registration, communications and questions related to the content of the comparative testing round.

The EU-RL GMFF is looking forward to your participation.

Yours sincerely,

Machin Kreysa

Head of Molecular Biology and Genomics Unit

Annex 7: Accompanying letter to shipment of samples





Ispra, 11 April 2014 JRC.DG.I.3/MBG/JK/mm/lv

NOTE FOR THE ATTENTION OF All Laboratories registered for the comparative test ILC-EURL-GMFF-CT-02/13

«Firstname» «Surname» «LCode» «Organisation» «Address» «Zip» «Town» «Country»

Subject: ILC-EURL-GMFF-CT-02/13, a comparative testing round to determine the GM content in two test materials of rice noodles, test items and tasks.

Dear «Title» «Surname»,

Thank you for participating in the above indicated ILC-EURL-GMFF-CT-02/13 comparative testing round. Please find in this parcel two test materials of rice noodles.

The parcel contains:

- 1. Two plastic containers each containing approximately 10 g of test item;
- 2. An "Acknowledgement of Reception" form.

Please check whether the plastic containers containing the test item remained undamaged during transport and return the signed "Acknowledgement of Reception" form by fax (+39 0332 786159) or by e-mail with scanned pdf. You should store the samples in a dark and cold place (not exceeding 18 °C).

Tasks

Participants should:

- 1. Perform species identification on each test item for: maize, soybean, oilseed rape and rice.
- Identify and quantify the GM event(s) detected.

The procedures used for detection/quantification of the detected GM events should resemble as closely as possible the ones that you use in routine sample analyses.

The results can be reported in mass/mass % or copy/copy % as outlined below:

mass/mass % = $\frac{\text{mass GM [g]}}{\text{Total mass [g]}} \times 100 \%$ $\text{copy/copy \%} = \frac{\text{GM DNA copy numbers [cp]}}{\text{Target taxon-specific DNA copy numbers [cp]}} \times 100 \%$

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E-mail: Joachim.kreysa@ec.europa.eu
http://ihcp.jrc.ec.europa.eu

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You can find the reporting website at https://web.jrc.ec.europa.eu/ilcReportingWeb. You need a personal password to access this webpage which is "Part_key". The system will guide you through the reporting procedure.

Please be aware that you are not allowed to report results both in mass/mass % and copy/copy %. The results of participants having submitted results in both measurement units will be discarded.

After entering all results, please complete the questionnaire. In the questionnaire, items bearing a question mark icon on the right-hand side contain additional information for the participant. In the reporting website clicking on the icon will give access to this information. Do not forget to save, submit and confirm when required to do so.

The pdf file of the questionnaire that you will receive by E-mail is intended as an aid in the laboratory.

Only results and answers to the questionnaire that are reported on-line on the reporting website https://web.jrc.ec.europa.eu/ilcReportingWeb will be accepted.

When reporting results, please note that fields carrying the indication "number" should only contain numeric values.

Directly after submitting your results and the questionnaire information on-line, you will be prompted to print the completed report form. Please sign the printed report form and return it to the EU-RL GMFF as scanned pdf by E-mail (mbg-comparative-testing@jrc.ec.europa.eu). Check your results carefully before submission, since this is your final confirmation. The EU-RL GMFF will not verify your data.

The deadline for submission of results is 30 May 2014. It will not be possible to submit your results after the deadline.

Please contact the functional mailbox mbg-comparative-testing@jrc.ec.europa.eu for all issues related to this comparative testing round.

Thank you very much for the collaboration in this comparative testing round.

Yours sincerely,

Joachim Kreysa Head of Unit

EU-RL GMFF: Comparative testing report

Annex 8: Confirmation of shipment

Dear Participant,

Your test parcels related to the eighth comparative testing round ILC-EURL-GMFF-CT-02/13 has left our premises today 15 April 2014 by TNT courier.

For your convenience, please find herewith the corresponding tracking number you could refer to in order to track the relevant materials on the Web:

25913 8945

The parcel with test items that you will receive should contain:

- Two plastic containers each containing approximately 10 g of test item;
- An "acknowledgement of reception" form, that should be returned to the **EU-RL GMFF** as scanned pdf by e-mail to mbg-comparative-testing@jrc.ec.europa.eu.
- An accompanying letter.

•

The accompanying letter contains your **personal password** for on-line submission of your results to the reporting website https://web.jrc.ec.europa.eu/ilcReportingWeb.

Your Lab Code (Lxx) is indicated in the accompanying letter as well in the "acknowledgement of reception" form in the upper right side of the page; please keep it for future uses in this CT round.

The deadline for submission of your results is 30 May 2014.

The questionnaire will be sent via separate e-mail.

Please contact only the functional mailbox mbg-comparative-testing@jrc.ec.europa.eu for any issue related to this comparative testing round.

Thank you for your collaboration.

Lorella Vidmar

On behalf of

The Comparative Testing staff



European Commission

DG Joint Research Centre

Institute for Health and Consumer Protection Unit I.3 Molecular Biology and Genomics TP 201 Via E. Fermi 2749

I-21027- Ispra (VA) Italy

Functional mailbox: mbq-comparative-testing@jrc.ec.europa.eu

EU-RL GMFF: Comparative testing report

Annex 9: Acknowledgement of receipt

DG IRC B		for GM food & Food
RC.I3.R71/EURL Date: 19/07/2011 Revision. 4	Acknowledgement of reception	Page 1/1
From:		Lab Code:
		39 0 332 78 6159
We have received the foll Two plastic containers each c	In good condition owing samples Ontaining approximately 10 g of rice noodles.	No
No information regarding party.	the sample(s) received and results of related testing may be	disclosed to any third
Comments:		
Date:	Visa:	
By signing this document the	participant agrees with the clause of non disclosure of information	on samples and results

FAX - Record for Quality System

Please send this document via EMAIL to:

mbg-comparative-testing@jrc.ec.europa.eu

This document is not a recognition of the quantity and/or quality of samples and reagents provided. This document will be used by EURL-GMFF only to confirm the reception of goods provided to participating laboratories in its Quality System. EURL-GMFF thanks you very much for your participation.

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European Commission

EUR 26863 EN - Joint Research Centre - Institute for Health and Consumer Protection

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Author(s): Niccolo Bassani, Adam Niedzwiecki, Angelo Collotta, Matteo Maretti, Wim Broothaerts, Marco Mazzara, Joachim Kreysa

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