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172-PT

PROFICIENCY TESTING 2012

INFECTIOUS BOVINE RHINOTRACHEITIS (IBR)

Detection of IBRgB- and IBRgE-specific antibodies in serum by Enzyme Linked Immunosorbent Assay (ELISA)

OPERATIONAL UNIT

COORDINATION OF VETERINARY DIAGNOSIS

EPIDEMIOLOGY AND RISK ASSESSMENT

(CVD-ERA)

DATE BEGIN PT: 04 JUNE 2012

DATE REPORT: 10 AUGUST 2012

I. Introduction

Details relevant to the proficiency test (PT) are available in the Procedure PRO/2.5/01 'Beheer van de proficiency testen op het CODA-CERVA-Ukkel/Gestion des essais d'aptitude au CODA-CERVA-Uccle', which is summarized in the 'Manual for the participant'.

II. Aim

The aim of this PT was to evaluate the ability of the participating laboratories to identify the absence or presence of IBRgB- and/or IBRgE-specific antibodies in bovine serum by ELISA.

III. Materials and methods

III.1. Conduct of diagnostic tests

In the framework of this PT, predefined reference serum samples must be analyzed by means of an ELISA test. The procedures for the ELISA tests must be fully described in the SOPs of the participating laboratories.

III.2. Reference samples

III.2.1. IBRgB reference samples

Replicates of 5 reference serum samples of bovine origin, either free from detectable IBRgB-specific antibodies (n = 2; coded 'PT2012IBRgBSERNS1' and 'PT2012IBRgBSERNS2') or containing detectable IBRgB-specific antibodies (n = 3; coded 'PT2012IBRgBSERPS1', 'PT2012IBRgBSERPS2' and 'PT2012IBRgBSERPS3'), were used. In total, 160 aliquots were distributed to 8 participating laboratories. All participants were given 4 aliquots of the 5 reference serum samples, i.e. 20 aliquots. The positions of the reference serum samples in the sent blocks were randomized for each participant (Table 4).

For each reference serum sample, a certificate containing the status of the sample (= 'golden standard') was made. The status of the reference serum samples was based on (i) the historical background of the animals and (ii) the results obtained by the HerdChek IBRgB antibody ELISA test from IDEXX, the indirect ELISA test from LSI (LSIVET serum IBR screening) and a seroneutralisation assay (SN) (pre-verification). The reference serum samples PT2012IBRgBSERNS1 and PT2012IBRgBSERNS2 were obtained from 2 animals from a Belgian I4-certified farm (IBR-free without vaccination). The reference serum sample PT2012IBRgBSERPS1 was a 1/128 dilution of a serum from a vaccinated but uninfected animal, whereas the reference serum samples PT2012IBRgBSERPS2 and PT2012IBRgBSERPS3 were a 1/32 and a 1/8 dilution, respectively, of 2 different sera from experimentally infected but non-vaccinated animals. For each reference serum sample, the same qualitative result was obtained with all test methods used. Taken together, the reference serum samples PT2012IBRgBSERNS1 and PT2012IBRgBSERNS2 were considered as negative sera, and the reference serum samples PT2012IBRgBSERPS1, PT2012IBRgBSERPS2 and PT2012IBRgBSERPS3 as weak positive sera in SN but strong positive sera in IBRgB ELISA.

After aliquoting the different reference serum samples, a homogeneity check was performed on 10 aliquots of each reference serum sample using the HerdChek IBRgB antibody ELISA test from IDEXX, hereby obtaining the same qualitative result for all 10 aliquots of the same reference serum sample. Consequently, all reference serum samples were considered as reliable samples in order to evaluate the ability of laboratories to correctly identify the absence or presence of IBRgB-specific antibodies in bovine serum. In addition, all reference serum samples were tested once after the PT in order to confirm their stability and status (post-verification) using the HerdChek IBRgB antibody ELISA test from IDEXX.

III.2.2. IBRgE reference samples

Replicates of 5 reference serum samples of bovine origin, either free from detectable IBRgE-specific antibodies (n = 3; coded 'PT2012IBRgESERNS1', 'PT2012IBRgESERNS2' and 'PT2012IBRgESERNS3') or containing detectable IBRgE-specific antibodies (n = 2; coded 'PT2012IBRgESERPS1' and 'PT2012IBRgESERPS2'), were used. In total, 140 aliquots were distributed to 7 participating laboratories. All participants were given 4 aliquots of the 5 reference serum samples, i.e.

20 aliquots. The positions of the reference serum samples in the sent blocks were randomized for each participant (Table 5).

For each reference serum sample, a certificate containing the status of the sample (= 'golden standard') was made. The status of the reference serum samples was based on (i) the historical background of the animals and (ii) the results obtained by the HerdChek IBRgE antibody ELISA test from IDEXX (pre-verification). The reference serum samples PT2012IBRgESERNS1 (=PT2012IBRgBSERNS1) and PT2012IBRgESERNS2 (=PT2012IBRgBSERNS2) were from 2 animals from a Belgian I4-certified farm (IBR-free without vaccination), whereas the reference serum sample PT2012IBRgESERNS3 was from a vaccinated but uninfected animal. The reference serum samples PT2012IBRgESERPS1 (=PT2012IBRgBSERPS2) and PT2012IBRgESERPS2 (=PT2012IBRgBSERPS3) were a 1/32 and a 1/8 dilution, respectively, of 2 different sera from experimentally infected but non-vaccinated animals. Taken together, the reference serum samples PT2012IBRgESERNS1, PT2012IBRgESERNS2 and PT2012IBRgESERNS3 were considered as negative sera, the reference serum sample PT2012IBRgESERPS1 as a weak positive serum and the reference serum sample PT2012IBRgESERPS2 as a strong positive serum in IBRgE ELISA.

After aliquoting the different reference serum samples, a homogeneity check was performed on 10 aliquots of each reference serum sample using the HerdChek IBRgE antibody ELISA test from IDEXX, hereby obtaining the same qualitative result for all 10 aliquots of the same reference serum sample. Consequently, all reference serum samples were considered as reliable samples in order to evaluate the ability of laboratories to correctly identify the absence or presence of IBRgE-specific antibodies in bovine serum. In addition, all reference serum samples were tested once after the PT in order to confirm their stability and status (post-verification) using the HerdChek IBRgE antibody ELISA test from IDEXX.

III.3. Classification of results, level of agreement and threshold for qualification

III.3.1. Classification of results

Results provided by the participating laboratories are categorized as *success* (positive result when the reference sample is truly positive, negative result when the reference sample is truly negative) or *failure* (positive result when the reference sample is truly negative, negative result when the reference sample is truly positive, non-interpretable result when the reference sample is truly negative or positive).

III.3.2. Level of agreement

The level of agreement achieved by a participating laboratory is expressed as the percentage *success* (i.e., the reported result matches with the assigned status) obtained for the 20 aliquots of reference serum samples used for either the IBRgB or the IBRgE PT.

III.3.3. Threshold for qualification

Following the procedure, a participating laboratory is only qualified if the level of agreement for the 20 aliquots of reference serum samples used for either PT is at least 90%.

IV. Results

For confidentiality reasons, the participating laboratories are quoted anonymously and the concordance table is safely kept at the operational unit CVD-ERA of CODA-CERVA.

IV.1. Transfer and start of the analyses of the reference samples

LAB1 until LAB7 participated in both the IBRgB and the IBRgE PT and received 40 aliquots of reference serum samples (20 for the IBRgB PT and 20 for the IBRgE PT). LAB8 only participated in the IBRgB PT and hence received 20 aliquots of reference serum samples. The reference serum samples were sent frozen (dry ice) to each of the participating laboratories by national or international courier on 4th of June 2012 (300 aliquots in total). LAB4, LAB5, LAB6 and LAB7 acknowledged receipt of the samples on the same day, whereas the other laboratories received the samples on 5th of June 2012. All participating laboratories confirmed that the reference serum samples were still frozen upon receipt. Analyses were performed between 4th and 12th of June 2012 (Table 1).

IV.2. Dates at which results were returned to the operational unit CVD-ERA

Results from the participating laboratories were submitted to the operational unit CVD-ERA between 6th and 18th of June 2012 (Table 1). LAB5 hereby exceeded the deadline of 15th of June 2012 for submission of the results.

Table 1. Overview of the dates on which (i) the reference serum samples were received and analyzed by the participating laboratories, and (ii) the obtained results were submitted to the operational unit CVD-ERA of CODA-CERVA.

Laboratory	Reference samples received	Start of analysis gB	Start of analysis gE	Submission of the results (Excel file)
LAB1	05/06/2012	07/06/2012	08/06/2012	14/06/2012
LAB2	05/06/2012	11/06/2012 (*)	11/06/2012 (*)	14/06/2012
LAB3	05/06/2012	11/06/2012 & 12/06/2012 (#)	11/06/2012 (°)	14/06/2012
LAB4	04/06/2012	07/06/2012	07/06/2012	08/06/2012
LAB5	04/06/2012	07/06/2012	07/06/2012	18/06/2012
LAB6	04/06/2012	07/06/2012	08/06/2012	14/06/2012
LAB7	04/06/2012	04/06/2012	04/06/2012	06/06/2012
LAB8	05/06/2012	08/06/2012 (*)	NA	11/06/2012

Legend: NA = not applicable; (*) = this laboratory performed both the short and the long incubation protocol for the same batch of the used ELISA kit; (#) = this laboratory tested 2 ELISA kits from different producers (same incubation protocol); (°) = this laboratory tested 2 different batches of the same ELISA kit (same incubation protocol)

IV.3. Compliance with the procedure

All participating laboratories have provided a duly dated and signed copy of the results.

IV.4. Qualitative data analysis

LAB2, LAB3 and LAB8 submitted 2 sets of results for the IBRgB and/or the IBRgE PT since they analysed the 20 aliquots of reference serum samples using ELISA kits from different producers, different batches of the same ELISA kit and/or different incubation protocols for the same batch of the used ELISA kit. In order to analyse the provided data, these 3 laboratories have been divided into different sublaboratories, namely: LAB2 into LAB2.1 (short incubation protocol for both the IBRgB and the IBRgE PT) and LAB2.2 (long incubation protocol for both the IBRgB and the IBRgE PT), LAB3 into LAB3.1 (kit1 for the IBRgB PT; batch1 for the IBRgE PT) and LAB3.2 (kit2 for the IBRgB PT; batch2 for the IBRgE PT), and LAB8 into LAB8.1 (long incubation protocol for the IBRgB PT) and LAB8.2 (short incubation protocol for the IBRgB PT).

IV.4.1. Level of agreement

Qualitative data analysis showed that:

- **(i)** For the detection of **IBRgB-specific antibodies**, all 8 participating laboratories provided qualitative results that were in full agreement with the true status of the reference serum samples (100% of agreement). Hereby, LAB3 used 2 different ELISA kits, whereas LAB2 and LAB8 performed both the short and the long incubation protocol for the same batch of the used ELISA kit (Table 2).
- **(ii)** For the detection of **IBRgE-specific antibodies**, 6 out of 7 participating laboratories provided qualitative results that were in full agreement with the true status of the reference serum samples (100% of agreement). Hereby, LAB3 used 2 different batches of the same ELISA kit. In contrast, LAB2 misclassified 3 aliquots performing the short incubation protocol (85% of agreement) and 1 aliquot performing the long incubation protocol (95% of agreement) of the same batch of the used ELISA kit (Table 3).

Table 2. Agreement between results generated by the participating laboratories (LABNR) and the status of the **IBRgB** reference serum samples assigned by the IBR reference laboratory of CODA-CERVA. All participating laboratories received 20 aliquots of IBRgB reference serum samples. Results are presented as absolute values and percentages (in parentheses).

	LABNR									
	1	**2.1**	**2.2**	**3.1**	**3.2**	**4**	**5**	**6**	**7**	**8.1**
Failure	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Success	20 (100.0)	20 (100.0)	20 (100.0)	20 (100.0)	20 (100.0)	20 (100.0)	20 (100.0)	20 (100.0)	20 (100.0)	20 (100.0)

	LABNR
	8.2
Failure	0 (0.0)
Success	20 (100.0)

Table 3. Agreement between results generated by the participating laboratories (LABNR) and the status of the **IBRgE** reference serum samples assigned by the IBR reference laboratory of CODA-CERVA. All participating laboratories received 20 aliquots of IBRgE reference serum samples. Results are presented as absolute values and percentages (in parentheses).

	LABNR								
	1	**2.1**	**2.2**	**3.1**	**3.2**	**4**	**5**	**6**	**7**
failure	0 (0.0)	3 (15.0)	1 (5.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
success	20 (100.0)	17 (85.0)	19 (95.0)	20 (100.0)	20 (100.0)	20 (100.0)	20 (100.0)	20 (100.0)	20 (100.0)

A quantitative data analysis (including box plots) is shown for educational purposes in Annex 1 and Annex 2.

IV.4.2. Variability among participating laboratories

Only a small variability between laboratories could be observed at the qualitative data level:

(i) For the detection of **IBRgB-specific antibodies** in reference serum samples, no variability between laboratories could be observed since all participants correctly identified all reference serum samples. Hereby, LAB3 used 2 different ELISA kits, whereas LAB2 and LAB8 performed both the short and the long incubation protocol for the same batch of the used ELISA kit.

(ii) For the detection of **IBRgE-specific antibodies** in reference serum samples, no variability between LAB1, LAB3, LAB4, LAB5, LAB6 and LAB7 could be observed since these participants correctly identified all reference serum samples. Hereby, LAB3 used 2 different batches of the same ELISA kit. In contrast, LAB2 misclassified 3 aliquots of the weak positive reference serum sample PT2012IBRgESERPS1 (3x NI instead of POS) performing the short incubation protocol and 1 aliquot of the same sample (NI instead of POS) performing the long incubation protocol.

For each participating laboratory, the obtained results and the assigned statuses for the reference serum samples are shown in Table 4 for the IBRgB PT and in Table 5 for the IBRgE PT.

Table 4. The responses (RESULT) of the participating laboratories (LABNR) with the identification of the **IBRgB** reference serum samples (SAMPLE), the positions of the IBRgB reference serum samples as placed in the block (LABPOSIT), and the status assigned by the IBR reference laboratory of CODA-CERVA (STATUS). NEG: negative; POS: positive.

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
1	1	1	PT2012IBRgBSERNS2	NEG	NEG	1
2	1	2	PT2012IBRgBSERNS1	NEG	NEG	1
3	1	3	PT2012IBRgBSERPS1	POS	POS	1
4	1	4	PT2012IBRgBSERPS1	POS	POS	1
5	1	5	PT2012IBRgBSERPS3	POS	POS	1
6	1	6	PT2012IBRgBSERPS2	POS	POS	1
7	1	7	PT2012IBRgBSERNS2	NEG	NEG	1
8	1	8	PT2012IBRgBSERPS2	POS	POS	1
9	1	9	PT2012IBRgBSERPS3	POS	POS	1
10	1	10	PT2012IBRgBSERNS1	NEG	NEG	1
11	1	11	PT2012IBRgBSERPS1	POS	POS	1
12	1	12	PT2012IBRgBSERPS3	POS	POS	1
13	1	13	PT2012IBRgBSERPS2	POS	POS	1
14	1	14	PT2012IBRgBSERPS3	POS	POS	1
15	1	15	PT2012IBRgBSERNS1	NEG	NEG	1
16	1	16	PT2012IBRgBSERPS2	POS	POS	1
17	1	17	PT2012IBRgBSERNS2	NEG	NEG	1
18	1	18	PT2012IBRgBSERPS1	POS	POS	1
19	1	19	PT2012IBRgBSERNS1	NEG	NEG	1
20	1	20	PT2012IBRgBSERNS2	NEG	NEG	1
21	2.1	1	PT2012IBRgBSERPS1	POS	POS	1
22	2.1	2	PT2012IBRgBSERPS3	POS	POS	1
23	2.1	3	PT2012IBRgBSERPS2	POS	POS	1
24	2.1	4	PT2012IBRgBSERNS2	NEG	NEG	1
25	2.1	5	PT2012IBRgBSERPS2	POS	POS	1
26	2.1	6	PT2012IBRgBSERPS3	POS	POS	1
27	2.1	7	PT2012IBRgBSERNS1	NEG	NEG	1
28	2.1	8	PT2012IBRgBSERPS1	POS	POS	1
29	2.1	9	PT2012IBRgBSERPS3	POS	POS	1
30	2.1	10	PT2012IBRgBSERPS2	POS	POS	1
31	2.1	11	PT2012IBRgBSERPS3	POS	POS	1
32	2.1	12	PT2012IBRgBSERNS1	NEG	NEG	1
33	2.1	13	PT2012IBRgBSERPS2	POS	POS	1
34	2.1	14	PT2012IBRgBSERNS2	NEG	NEG	1
35	2.1	15	PT2012IBRgBSERPS1	POS	POS	1
36	2.1	16	PT2012IBRgBSERNS1	NEG	NEG	1
37	2.1	17	PT2012IBRgBSERNS2	NEG	NEG	1
38	2.1	18	PT2012IBRgBSERNS2	NEG	NEG	1
39	2.1	19	PT2012IBRgBSERNS1	NEG	NEG	1
40	2.1	20	PT2012IBRgBSERPS1	POS	POS	1

(Table 4 - CONTINUED)

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
41	2.2	1	PT2012IBRgBSERPS1	POS	POS	1
42	2.2	2	PT2012IBRgBSERPS3	POS	POS	1
43	2.2	3	PT2012IBRgBSERPS2	POS	POS	1
44	2.2	4	PT2012IBRgBSERNS2	NEG	NEG	1
45	2.2	5	PT2012IBRgBSERPS2	POS	POS	1
46	2.2	6	PT2012IBRgBSERPS3	POS	POS	1
47	2.2	7	PT2012IBRgBSERNS1	NEG	NEG	1
48	2.2	8	PT2012IBRgBSERPS1	POS	POS	1
49	2.2	9	PT2012IBRgBSERPS3	POS	POS	1
50	2.2	10	PT2012IBRgBSERPS2	POS	POS	1
51	2.2	11	PT2012IBRgBSERPS3	POS	POS	1
52	2.2	12	PT2012IBRgBSERNS1	NEG	NEG	1
53	2.2	13	PT2012IBRgBSERPS2	POS	POS	1
54	2.2	14	PT2012IBRgBSERNS2	NEG	NEG	1
55	2.2	15	PT2012IBRgBSERPS1	POS	POS	1
56	2.2	16	PT2012IBRgBSERNS1	NEG	NEG	1
57	2.2	17	PT2012IBRgBSERNS2	NEG	NEG	1
58	2.2	18	PT2012IBRgBSERNS2	NEG	NEG	1
59	2.2	19	PT2012IBRgBSERNS1	NEG	NEG	1
60	2.2	20	PT2012IBRgBSERPS1	POS	POS	1
61	3.1	1	PT2012IBRgBSERNS2	NEG	NEG	1
62	3.1	2	PT2012IBRgBSERPS2	POS	POS	1
63	3.1	3	PT2012IBRgBSERPS3	POS	POS	1
64	3.1	4	PT2012IBRgBSERNS1	NEG	NEG	1
65	3.1	5	PT2012IBRgBSERPS1	POS	POS	1
66	3.1	6	PT2012IBRgBSERPS3	POS	POS	1
67	3.1	7	PT2012IBRgBSERPS2	POS	POS	1
68	3.1	8	PT2012IBRgBSERPS3	POS	POS	1
69	3.1	9	PT2012IBRgBSERNS1	NEG	NEG	1
70	3.1	10	PT2012IBRgBSERPS2	POS	POS	1
71	3.1	11	PT2012IBRgBSERNS2	NEG	NEG	1
72	3.1	12	PT2012IBRgBSERPS1	POS	POS	1
73	3.1	13	PT2012IBRgBSERNS1	NEG	NEG	1
74	3.1	14	PT2012IBRgBSERNS2	NEG	NEG	1
75	3.1	15	PT2012IBRgBSERNS2	NEG	NEG	1
76	3.1	16	PT2012IBRgBSERNS1	NEG	NEG	1
77	3.1	17	PT2012IBRgBSERPS1	POS	POS	1
78	3.1	18	PT2012IBRgBSERPS1	POS	POS	1
79	3.1	19	PT2012IBRgBSERPS3	POS	POS	1
80	3.1	20	PT2012IBRgBSERPS2	POS	POS	1

(Table 4 - CONTINUED)

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
81	3.2	1	PT2012IBRgBSERNS2	NEG	NEG	1
82	3.2	2	PT2012IBRgBSERPS2	POS	POS	1
83	3.2	3	PT2012IBRgBSERPS3	POS	POS	1
84	3.2	4	PT2012IBRgBSERNS1	NEG	NEG	1
85	3.2	5	PT2012IBRgBSERPS1	POS	POS	1
86	3.2	6	PT2012IBRgBSERPS3	POS	POS	1
87	3.2	7	PT2012IBRgBSERPS2	POS	POS	1
88	3.2	8	PT2012IBRgBSERPS3	POS	POS	1
89	3.2	9	PT2012IBRgBSERNS1	NEG	NEG	1
90	3.2	10	PT2012IBRgBSERPS2	POS	POS	1
91	3.2	11	PT2012IBRgBSERNS2	NEG	NEG	1
92	3.2	12	PT2012IBRgBSERPS1	POS	POS	1
93	3.2	13	PT2012IBRgBSERNS1	NEG	NEG	1
94	3.2	14	PT2012IBRgBSERNS2	NEG	NEG	1
95	3.2	15	PT2012IBRgBSERNS2	NEG	NEG	1
96	3.2	16	PT2012IBRgBSERNS1	NEG	NEG	1
97	3.2	17	PT2012IBRgBSERPS1	POS	POS	1
98	3.2	18	PT2012IBRgBSERPS1	POS	POS	1
99	3.2	19	PT2012IBRgBSERPS3	POS	POS	1
100	3.2	20	PT2012IBRgBSERPS2	POS	POS	1
101	4	1	PT2012IBRgBSERNS1	NEG	NEG	1
102	4	2	PT2012IBRgBSERPS1	POS	POS	1
103	4	3	PT2012IBRgBSERPS3	POS	POS	1
104	4	4	PT2012IBRgBSERPS2	POS	POS	1
105	4	5	PT2012IBRgBSERPS3	POS	POS	1
106	4	6	PT2012IBRgBSERNS1	NEG	NEG	1
107	4	7	PT2012IBRgBSERPS2	POS	POS	1
108	4	8	PT2012IBRgBSERNS2	NEG	NEG	1
109	4	9	PT2012IBRgBSERPS1	POS	POS	1
110	4	10	PT2012IBRgBSERNS1	NEG	NEG	1
111	4	11	PT2012IBRgBSERNS2	NEG	NEG	1
112	4	12	PT2012IBRgBSERNS2	NEG	NEG	1
113	4	13	PT2012IBRgBSERNS1	NEG	NEG	1
114	4	14	PT2012IBRgBSERPS1	POS	POS	1
115	4	15	PT2012IBRgBSERPS1	POS	POS	1
116	4	16	PT2012IBRgBSERPS3	POS	POS	1
117	4	17	PT2012IBRgBSERPS2	POS	POS	1
118	4	18	PT2012IBRgBSERNS2	NEG	NEG	1
119	4	19	PT2012IBRgBSERPS2	POS	POS	1
120	4	20	PT2012IBRgBSERPS3	POS	POS	1

(Table 4 - CONTINUED)

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
121	5	1	PT2012IBRgBSERPS2	POS	POS	1
122	5	2	PT2012IBRgBSERPS3	POS	POS	1
123	5	3	PT2012IBRgBSERNS1	NEG	NEG	1
124	5	4	PT2012IBRgBSERPS2	POS	POS	1
125	5	5	PT2012IBRgBSERNS2	NEG	NEG	1
126	5	6	PT2012IBRgBSERPS1	POS	POS	1
127	5	7	PT2012IBRgBSERNS1	NEG	NEG	1
128	5	8	PT2012IBRgBSERNS2	NEG	NEG	1
129	5	9	PT2012IBRgBSERNS2	NEG	NEG	1
130	5	10	PT2012IBRgBSERNS1	NEG	NEG	1
131	5	11	PT2012IBRgBSERPS1	POS	POS	1
132	5	12	PT2012IBRgBSERPS1	POS	POS	1
133	5	13	PT2012IBRgBSERPS3	POS	POS	1
134	5	14	PT2012IBRgBSERPS2	POS	POS	1
135	5	15	PT2012IBRgBSERNS2	NEG	NEG	1
136	5	16	PT2012IBRgBSERPS2	POS	POS	1
137	5	17	PT2012IBRgBSERPS3	POS	POS	1
138	5	18	PT2012IBRgBSERNS1	NEG	NEG	1
139	5	19	PT2012IBRgBSERPS1	POS	POS	1
140	5	20	PT2012IBRgBSERPS3	POS	POS	1
141	6	1	PT2012IBRgBSERPS2	POS	POS	1
142	6	2	PT2012IBRgBSERNS2	NEG	NEG	1
143	6	3	PT2012IBRgBSERPS1	POS	POS	1
144	6	4	PT2012IBRgBSERNS1	NEG	NEG	1
145	6	5	PT2012IBRgBSERNS2	NEG	NEG	1
146	6	6	PT2012IBRgBSERNS2	NEG	NEG	1
147	6	7	PT2012IBRgBSERNS1	NEG	NEG	1
148	6	8	PT2012IBRgBSERPS1	POS	POS	1
149	6	9	PT2012IBRgBSERPS1	POS	POS	1
150	6	10	PT2012IBRgBSERPS3	POS	POS	1
151	6	11	PT2012IBRgBSERPS2	POS	POS	1
152	6	12	PT2012IBRgBSERNS2	NEG	NEG	1
153	6	13	PT2012IBRgBSERPS2	POS	POS	1
154	6	14	PT2012IBRgBSERPS3	POS	POS	1
155	6	15	PT2012IBRgBSERNS1	NEG	NEG	1
156	6	16	PT2012IBRgBSERPS1	POS	POS	1
157	6	17	PT2012IBRgBSERPS3	POS	POS	1
158	6	18	PT2012IBRgBSERPS2	POS	POS	1
159	6	19	PT2012IBRgBSERPS3	POS	POS	1
160	6	20	PT2012IBRgBSERNS1	NEG	NEG	1

(Table 4 - CONTINUED)

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
161	7	1	PT2012IBRgBSERNS1	NEG	NEG	1
162	7	2	PT2012IBRgBSERNS2	NEG	NEG	1
163	7	3	PT2012IBRgBSERNS2	NEG	NEG	1
164	7	4	PT2012IBRgBSERNS1	NEG	NEG	1
165	7	5	PT2012IBRgBSERPS1	POS	POS	1
166	7	6	PT2012IBRgBSERPS1	POS	POS	1
167	7	7	PT2012IBRgBSERPS3	POS	POS	1
168	7	8	PT2012IBRgBSERPS2	POS	POS	1
169	7	9	PT2012IBRgBSERNS2	NEG	NEG	1
170	7	10	PT2012IBRgBSERPS2	POS	POS	1
171	7	11	PT2012IBRgBSERPS3	POS	POS	1
172	7	12	PT2012IBRgBSERNS1	NEG	NEG	1
173	7	13	PT2012IBRgBSERPS1	POS	POS	1
174	7	14	PT2012IBRgBSERPS3	POS	POS	1
175	7	15	PT2012IBRgBSERPS2	POS	POS	1
176	7	16	PT2012IBRgBSERPS3	POS	POS	1
177	7	17	PT2012IBRgBSERNS1	NEG	NEG	1
178	7	18	PT2012IBRgBSERPS2	POS	POS	1
179	7	19	PT2012IBRgBSERNS2	NEG	NEG	1
180	7	20	PT2012IBRgBSERPS1	POS	POS	1
181	8.1	1	PT2012IBRgBSERNS1	NEG	NEG	1
182	8.1	2	PT2012IBRgBSERPS1	POS	POS	1
183	8.1	3	PT2012IBRgBSERPS1	POS	POS	1
184	8.1	4	PT2012IBRgBSERPS3	POS	POS	1
185	8.1	5	PT2012IBRgBSERPS2	POS	POS	1
186	8.1	6	PT2012IBRgBSERNS2	NEG	NEG	1
187	8.1	7	PT2012IBRgBSERPS2	POS	POS	1
188	8.1	8	PT2012IBRgBSERPS3	POS	POS	1
189	8.1	9	PT2012IBRgBSERNS1	NEG	NEG	1
190	8.1	10	PT2012IBRgBSERPS1	POS	POS	1
191	8.1	11	PT2012IBRgBSERPS3	POS	POS	1
192	8.1	12	PT2012IBRgBSERPS2	POS	POS	1
193	8.1	13	PT2012IBRgBSERPS3	POS	POS	1
194	8.1	14	PT2012IBRgBSERNS1	NEG	NEG	1
195	8.1	15	PT2012IBRgBSERPS2	POS	POS	1
196	8.1	16	PT2012IBRgBSERNS2	NEG	NEG	1
197	8.1	17	PT2012IBRgBSERPS1	POS	POS	1
198	8.1	18	PT2012IBRgBSERNS1	NEG	NEG	1
199	8.1	19	PT2012IBRgBSERNS2	NEG	NEG	1
200	8.1	20	PT2012IBRgBSERNS2	NEG	NEG	1

(Table 4 - CONTINUED)

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
201	8.2	1	PT2012IBRgBSERNS1	NEG	NEG	1
202	8.2	2	PT2012IBRgBSERPS1	POS	POS	1
203	8.2	3	PT2012IBRgBSERPS1	POS	POS	1
204	8.2	4	PT2012IBRgBSERPS3	POS	POS	1
205	8.2	5	PT2012IBRgBSERPS2	POS	POS	1
206	8.2	6	PT2012IBRgBSERNS2	NEG	NEG	1
207	8.2	7	PT2012IBRgBSERPS2	POS	POS	1
208	8.2	8	PT2012IBRgBSERPS3	POS	POS	1
209	8.2	9	PT2012IBRgBSERNS1	NEG	NEG	1
210	8.2	10	PT2012IBRgBSERPS1	POS	POS	1
211	8.2	11	PT2012IBRgBSERPS3	POS	POS	1
212	8.2	12	PT2012IBRgBSERPS2	POS	POS	1
213	8.2	13	PT2012IBRgBSERPS3	POS	POS	1
214	8.2	14	PT2012IBRgBSERNS1	NEG	NEG	1
215	8.2	15	PT2012IBRgBSERPS2	POS	POS	1
216	8.2	16	PT2012IBRgBSERNS2	NEG	NEG	1
217	8.2	17	PT2012IBRgBSERPS1	POS	POS	1
218	8.2	18	PT2012IBRgBSERNS1	NEG	NEG	1
219	8.2	19	PT2012IBRgBSERNS2	NEG	NEG	1
220	8.2	20	PT2012IBRgBSERNS2	NEG	NEG	1

Table 5. The responses (RESULT) of the participating laboratories (LABNR) with the identification of the **IBRgE** reference serum samples (SAMPLE), the positions of the IBRgE reference serum samples as placed in the block (LABPOSIT), and the status assigned by the IBR reference laboratory of CODA-CERVA (STATUS). NEG: negative; POS: positive; NI: non-interpretable.

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
1	1	1	PT2012IBRgESERNS2	NEG	NEG	1
2	1	2	PT2012IBRgESERPS2	POS	POS	1
3	1	3	PT2012IBRgESERNS3	NEG	NEG	1
4	1	4	PT2012IBRgESERNS1	NEG	NEG	1
5	1	5	PT2012IBRgESERPS1	POS	POS	1
6	1	6	PT2012IBRgESERNS3	NEG	NEG	1
7	1	7	PT2012IBRgESERPS2	POS	POS	1
8	1	8	PT2012IBRgESERNS3	NEG	NEG	1
9	1	9	PT2012IBRgESERNS1	NEG	NEG	1
10	1	10	PT2012IBRgESERPS2	POS	POS	1
11	1	11	PT2012IBRgESERNS2	NEG	NEG	1
12	1	12	PT2012IBRgESERPS1	POS	POS	1
13	1	13	PT2012IBRgESERNS1	NEG	NEG	1
14	1	14	PT2012IBRgESERNS2	NEG	NEG	1
15	1	15	PT2012IBRgESERNS2	NEG	NEG	1
16	1	16	PT2012IBRgESERNS1	NEG	NEG	1
17	1	17	PT2012IBRgESERPS1	POS	POS	1
18	1	18	PT2012IBRgESERPS1	POS	POS	1
19	1	19	PT2012IBRgESERNS3	NEG	NEG	1
20	1	20	PT2012IBRgESERPS2	POS	POS	1
21	2.1	1	PT2012IBRgESERNS1	NEG	NEG	1
22	2.1	2	PT2012IBRgESERPS1	**POS**	**NI**	**0**
23	2.1	3	PT2012IBRgESERNS3	NEG	NEG	1
24	2.1	4	PT2012IBRgESERPS2	POS	POS	1
25	2.1	5	PT2012IBRgESERNS3	NEG	NEG	1
26	2.1	6	PT2012IBRgESERNS1	NEG	NEG	1
27	2.1	7	PT2012IBRgESERPS2	POS	POS	1
28	2.1	8	PT2012IBRgESERNS2	NEG	NEG	1
29	2.1	9	PT2012IBRgESERPS1	**POS**	**NI**	**0**
30	2.1	10	PT2012IBRgESERNS1	NEG	NEG	1
31	2.1	11	PT2012IBRgESERNS2	NEG	NEG	1
32	2.1	12	PT2012IBRgESERNS2	NEG	NEG	1
33	2.1	13	PT2012IBRgESERNS1	NEG	NEG	1
34	2.1	14	PT2012IBRgESERPS1	POS	POS	1
35	2.1	15	PT2012IBRgESERPS1	**POS**	**NI**	**0**
36	2.1	16	PT2012IBRgESERNS3	NEG	NEG	1
37	2.1	17	PT2012IBRgESERPS2	POS	POS	1
38	2.1	18	PT2012IBRgESERNS2	NEG	NEG	1
39	2.1	19	PT2012IBRgESERPS2	POS	POS	1
40	2.1	20	PT2012IBRgESERNS3	NEG	NEG	1

(Table 5 - CONTINUED)

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
41	2.2	1	PT2012IBRgESERNS1	NEG	NEG	1
42	2.2	2	PT2012IBRgESERPS1	POS	POS	1
43	2.2	3	PT2012IBRgESERNS3	NEG	NEG	1
44	2.2	4	PT2012IBRgESERPS2	POS	POS	1
45	2.2	5	PT2012IBRgESERNS3	NEG	NEG	1
46	2.2	6	PT2012IBRgESERNS1	NEG	NEG	1
47	2.2	7	PT2012IBRgESERPS2	POS	POS	1
48	2.2	8	PT2012IBRgESERNS2	NEG	NEG	1
49	2.2	9	PT2012IBRgESERPS1	POS	POS	1
50	2.2	10	PT2012IBRgESERNS1	NEG	NEG	1
51	2.2	11	PT2012IBRgESERNS2	NEG	NEG	1
52	2.2	12	PT2012IBRgESERNS2	NEG	NEG	1
53	2.2	13	PT2012IBRgESERNS1	NEG	NEG	1
54	2.2	14	PT2012IBRgESERPS1	POS	POS	1
55	2.2	15	PT2012IBRgESERPS1	**POS**	**NI**	**0**
56	2.2	16	PT2012IBRgESERNS3	NEG	NEG	1
57	2.2	17	PT2012IBRgESERPS2	POS	POS	1
58	2.2	18	PT2012IBRgESERNS2	NEG	NEG	1
59	2.2	19	PT2012IBRgESERPS2	POS	POS	1
60	2.2	20	PT2012IBRgESERNS3	NEG	NEG	1
61	3.1	1	PT2012IBRgESERPS2	POS	POS	1
62	3.1	2	PT2012IBRgESERNS3	NEG	NEG	1
63	3.1	3	PT2012IBRgESERNS1	NEG	NEG	1
64	3.1	4	PT2012IBRgESERPS2	POS	POS	1
65	3.1	5	PT2012IBRgESERNS2	NEG	NEG	1
66	3.1	6	PT2012IBRgESERPS1	POS	POS	1
67	3.1	7	PT2012IBRgESERNS1	NEG	NEG	1
68	3.1	8	PT2012IBRgESERNS2	NEG	NEG	1
69	3.1	9	PT2012IBRgESERNS2	NEG	NEG	1
70	3.1	10	PT2012IBRgESERNS1	NEG	NEG	1
71	3.1	11	PT2012IBRgESERPS1	POS	POS	1
72	3.1	12	PT2012IBRgESERPS1	POS	POS	1
73	3.1	13	PT2012IBRgESERNS3	NEG	NEG	1
74	3.1	14	PT2012IBRgESERPS2	POS	POS	1
75	3.1	15	PT2012IBRgESERNS2	NEG	NEG	1
76	3.1	16	PT2012IBRgESERPS2	POS	POS	1
77	3.1	17	PT2012IBRgESERNS3	NEG	NEG	1
78	3.1	18	PT2012IBRgESERNS1	NEG	NEG	1
79	3.1	19	PT2012IBRgESERPS1	POS	POS	1
80	3.1	20	PT2012IBRgESERNS3	NEG	NEG	1

(Table 5 - CONTINUED)

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
81	3.2	1	PT2012IBRgESERPS2	POS	POS	1
82	3.2	2	PT2012IBRgESERNS3	NEG	NEG	1
83	3.2	3	PT2012IBRgESERNS1	NEG	NEG	1
84	3.2	4	PT2012IBRgESERPS2	POS	POS	1
85	3.2	5	PT2012IBRgESERNS2	NEG	NEG	1
86	3.2	6	PT2012IBRgESERPS1	POS	POS	1
87	3.2	7	PT2012IBRgESERNS1	NEG	NEG	1
88	3.2	8	PT2012IBRgESERNS2	NEG	NEG	1
89	3.2	9	PT2012IBRgESERNS2	NEG	NEG	1
90	3.2	10	PT2012IBRgESERNS1	NEG	NEG	1
91	3.2	11	PT2012IBRgESERPS1	POS	POS	1
92	3.2	12	PT2012IBRgESERPS1	POS	POS	1
93	3.2	13	PT2012IBRgESERNS3	NEG	NEG	1
94	3.2	14	PT2012IBRgESERPS2	POS	POS	1
95	3.2	15	PT2012IBRgESERNS2	NEG	NEG	1
96	3.2	16	PT2012IBRgESERPS2	POS	POS	1
97	3.2	17	PT2012IBRgESERNS3	NEG	NEG	1
98	3.2	18	PT2012IBRgESERNS1	NEG	NEG	1
99	3.2	19	PT2012IBRgESERPS1	POS	POS	1
100	3.2	20	PT2012IBRgESERNS3	NEG	NEG	1
101	4	1	PT2012IBRgESERPS2	POS	POS	1
102	4	2	PT2012IBRgESERNS2	NEG	NEG	1
103	4	3	PT2012IBRgESERPS1	POS	POS	1
104	4	4	PT2012IBRgESERNS1	NEG	NEG	1
105	4	5	PT2012IBRgESERNS2	NEG	NEG	1
106	4	6	PT2012IBRgESERNS2	NEG	NEG	1
107	4	7	PT2012IBRgESERNS1	NEG	NEG	1
108	4	8	PT2012IBRgESERPS1	POS	POS	1
109	4	9	PT2012IBRgESERPS1	POS	POS	1
110	4	10	PT2012IBRgESERNS3	NEG	NEG	1
111	4	11	PT2012IBRgESERPS2	POS	POS	1
112	4	12	PT2012IBRgESERNS2	NEG	NEG	1
113	4	13	PT2012IBRgESERPS2	POS	POS	1
114	4	14	PT2012IBRgESERNS3	NEG	NEG	1
115	4	15	PT2012IBRgESERNS1	NEG	NEG	1
116	4	16	PT2012IBRgESERPS1	POS	POS	1
117	4	17	PT2012IBRgESERNS3	NEG	NEG	1
118	4	18	PT2012IBRgESERPS2	POS	POS	1
119	4	19	PT2012IBRgESERNS3	NEG	NEG	1
120	4	20	PT2012IBRgESERNS1	NEG	NEG	1

(Table 5 - CONTINUED)

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
121	5	1	PT2012IBRgESERNS1	NEG	NEG	1
122	5	2	PT2012IBRgESERNS2	NEG	NEG	1
123	5	3	PT2012IBRgESERNS2	NEG	NEG	1
124	5	4	PT2012IBRgESERNS1	NEG	NEG	1
125	5	5	PT2012IBRgESERPS1	POS	POS	1
126	5	6	PT2012IBRgESERPS1	POS	POS	1
127	5	7	PT2012IBRgESERNS3	NEG	NEG	1
128	5	8	PT2012IBRgESERPS2	POS	POS	1
129	5	9	PT2012IBRgESERNS2	NEG	NEG	1
130	5	10	PT2012IBRgESERPS2	POS	POS	1
131	5	11	PT2012IBRgESERNS3	NEG	NEG	1
132	5	12	PT2012IBRgESERNS1	NEG	NEG	1
133	5	13	PT2012IBRgESERPS1	POS	POS	1
134	5	14	PT2012IBRgESERNS3	NEG	NEG	1
135	5	15	PT2012IBRgESERPS2	POS	POS	1
136	5	16	PT2012IBRgESERNS3	NEG	NEG	1
137	5	17	PT2012IBRgESERNS1	NEG	NEG	1
138	5	18	PT2012IBRgESERPS2	POS	POS	1
139	5	19	PT2012IBRgESERNS2	NEG	NEG	1
140	5	20	PT2012IBRgESERPS1	POS	POS	1
141	6	1	PT2012IBRgESERNS1	NEG	NEG	1
142	6	2	PT2012IBRgESERPS1	POS	POS	1
143	6	3	PT2012IBRgESERPS1	POS	POS	1
144	6	4	PT2012IBRgESERNS3	NEG	NEG	1
145	6	5	PT2012IBRgESERPS2	POS	POS	1
146	6	6	PT2012IBRgESERNS2	NEG	NEG	1
147	6	7	PT2012IBRgESERPS2	POS	POS	1
148	6	8	PT2012IBRgESERNS3	NEG	NEG	1
149	6	9	PT2012IBRgESERNS1	NEG	NEG	1
150	6	10	PT2012IBRgESERPS1	POS	POS	1
151	6	11	PT2012IBRgESERNS3	NEG	NEG	1
152	6	12	PT2012IBRgESERPS2	POS	POS	1
153	6	13	PT2012IBRgESERNS3	NEG	NEG	1
154	6	14	PT2012IBRgESERNS1	NEG	NEG	1
155	6	15	PT2012IBRgESERPS2	POS	POS	1
156	6	16	PT2012IBRgESERNS2	NEG	NEG	1
157	6	17	PT2012IBRgESERPS1	POS	POS	1
158	6	18	PT2012IBRgESERNS1	NEG	NEG	1
159	6	19	PT2012IBRgESERNS2	NEG	NEG	1
160	6	20	PT2012IBRgESERNS2	NEG	NEG	1

(Table 5 - CONTINUED)

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
161	7	1	PT2012IBRgESERNS3	NEG	NEG	1
162	7	2	PT2012IBRgESERPS2	POS	POS	1
163	7	3	PT2012IBRgESERNS2	NEG	NEG	1
164	7	4	PT2012IBRgESERPS2	POS	POS	1
165	7	5	PT2012IBRgESERNS3	NEG	NEG	1
166	7	6	PT2012IBRgESERNS1	NEG	NEG	1
167	7	7	PT2012IBRgESERPS1	POS	POS	1
168	7	8	PT2012IBRgESERNS3	NEG	NEG	1
169	7	9	PT2012IBRgESERPS2	POS	POS	1
170	7	10	PT2012IBRgESERNS3	NEG	NEG	1
171	7	11	PT2012IBRgESERNS1	NEG	NEG	1
172	7	12	PT2012IBRgESERPS2	POS	POS	1
173	7	13	PT2012IBRgESERNS2	NEG	NEG	1
174	7	14	PT2012IBRgESERPS1	POS	POS	1
175	7	15	PT2012IBRgESERNS1	NEG	NEG	1
176	7	16	PT2012IBRgESERNS2	NEG	NEG	1
177	7	17	PT2012IBRgESERNS2	NEG	NEG	1
178	7	18	PT2012IBRgESERNS1	NEG	NEG	1
179	7	19	PT2012IBRgESERPS1	POS	POS	1
180	7	20	PT2012IBRgESERPS1	POS	POS	1

V. Discussion

The purpose of this PT was to assess performances of the participating laboratories when analyzing reference serum samples of bovine origin for the detection of IBRgB- and/or IBRgE-specific antibodies by ELISA.

For the detection of IBRgB-specific antibodies in reference serum samples, all 8 participating laboratories provided qualitative results that were in full agreement with the true status of the reference serum samples. Both LAB2 and LAB8 obtained identical qualitative results performing the short and the long incubation protocol for the same batch of the used IBRgB ELISA kit (2 different producers), while LAB3 obtained identical qualitative results performing the long incubation protocol of 2 IBRgB ELISA kits from different producers (Table 2 and Table 4).
The IBRgB participating laboratories used ELISA kits from 4 different producers as well as different batches from the same ELISA kit: IDEXX (3 batches: 1271, Y271 and Z211), LDL (1 batch: 12-01.1BHV), LSI (1 batch: 5-IBRG-001) and Synbiotics (1 batch: 11SIBR1BN210). LAB1, LAB3.2, LAB4, LAB5, LAB6 and LAB7 used an IBRgB ELISA kit from the same producer. Hereby, LAB1 was the only participant that performed the short incubation protocol.

For the detection of IBRgE-specific antibodies in reference serum samples, 6 out of 7 participating laboratories provided qualitative results that were in full agreement with the true status of the reference serum samples. LAB3 obtained identical qualitative results using 2 different batches of the same IBRgE ELISA kit. In contrast, LAB2 obtained different qualitative results performing the short and the long incubation protocol for the same batch of the used IBRgE ELISA kit: 3 aliquots of the weak positive reference serum sample PT2012IBRgESERPS1 were misclassified performing the short incubation protocol (85% of agreement), whereas 1 aliquot of the same sample was misclassified performing the long incubation protocol (95% of agreement) (Table 3 and Table 5).
The IBRgE participating laboratories used ELISA kits from 2 different producers as well as different batches from the same ELISA kit: IDEXX (4 batches: FG470R, MG291, CH644 and MG303) and Hipra (1 batch: CGE.24YH). LAB1, LAB3.1, LAB3.2, LAB4, LAB5, LAB6 and LAB7 used an IBRgE ELISA kit from the same producer. Hereby, all participants performed the long incubation protocol.

VI. Conclusions

According to the procedure currently in force, the performance of a participating laboratory is satisfactory if at least 90% of the results provided by this laboratory is in agreement with the status of the reference serum samples assigned by the IBR reference laboratory of CODA-CERVA (see III.3.3.). Consequently, all participants in the IBRgB PT achieved a satisfactory performance for the detection of IBRgB-specific antibodies in reference serum samples with all ELISA kits or incubation protocols used, whereas 6 out of 7 laboratories that participated in the IBRgE PT achieved a satisfactory performance for the detection of IBRgE-specific antibodies in reference serum samples with all batches used. For the IBRgE PT, LAB2 did reach the required 90% of agreement for the long incubation protocol, but not for the short incubation protocol of the used ELISA kit.

Head CVD-ERA
Yves Van der Stede

Appendix

Names of the participating laboratories

Agence nationale de sécurité sanitaire de l'alimentation, de l'environnement et du travail (ANSES) (Sophia-Antipolis, France)

Association Régionale de Santé et d'Identification Animales (ARSIA) (Ciney, Belgium)

Dierengezondheidszorg Vlaanderen (DGZ) (Torhout, Belgium)

Friedrich-Loeffler-Institut (FLI) (Greifswald-Insel-Riems, Germany)

Laboratoire de Médecine Vétérinaire de l'Etat (LMVE) (Grand Duchy of Luxemburg)

Laboratoire Service International (LSI) (Lissieu, France)

Synbiotics Europe (Lyon, France)

Veterinary and Agrochemical Research Center (CODA-CERVA) (Ukkel, Belgium)

Annex 1: Quantitative data analysis

Besides qualitative data analysis (positive, negative or non-interpretable result), also quantitative data analysis was performed using the statistical software programs SAS 9.2. (summary statistics) and R (box plots). All quantitative data analyses were performed on the normalized data, namely the percentages blocking calculated according to the instructions for this PT: [1- (OD_{Sample} / mean $OD_{Negative\ Kit\ Controls}$)] x 100.

The quantitative data analysis in this report was not used to evaluate the participants in this PT, but should only be considered as educational information for the participants in order to evaluate their performance and/or to standardize their different diagnostic tests.

I. Box plots

Box plots of the percentages blocking (calculated according to the instructions for this PT) per reference serum sample and per participating (sub)laboratory were made using the statistical software R. Box plots for the (sub)laboratories participating in the IBRgB PT and the IBRgE PT are shown in Figure 1 and Figure 2, respectively.

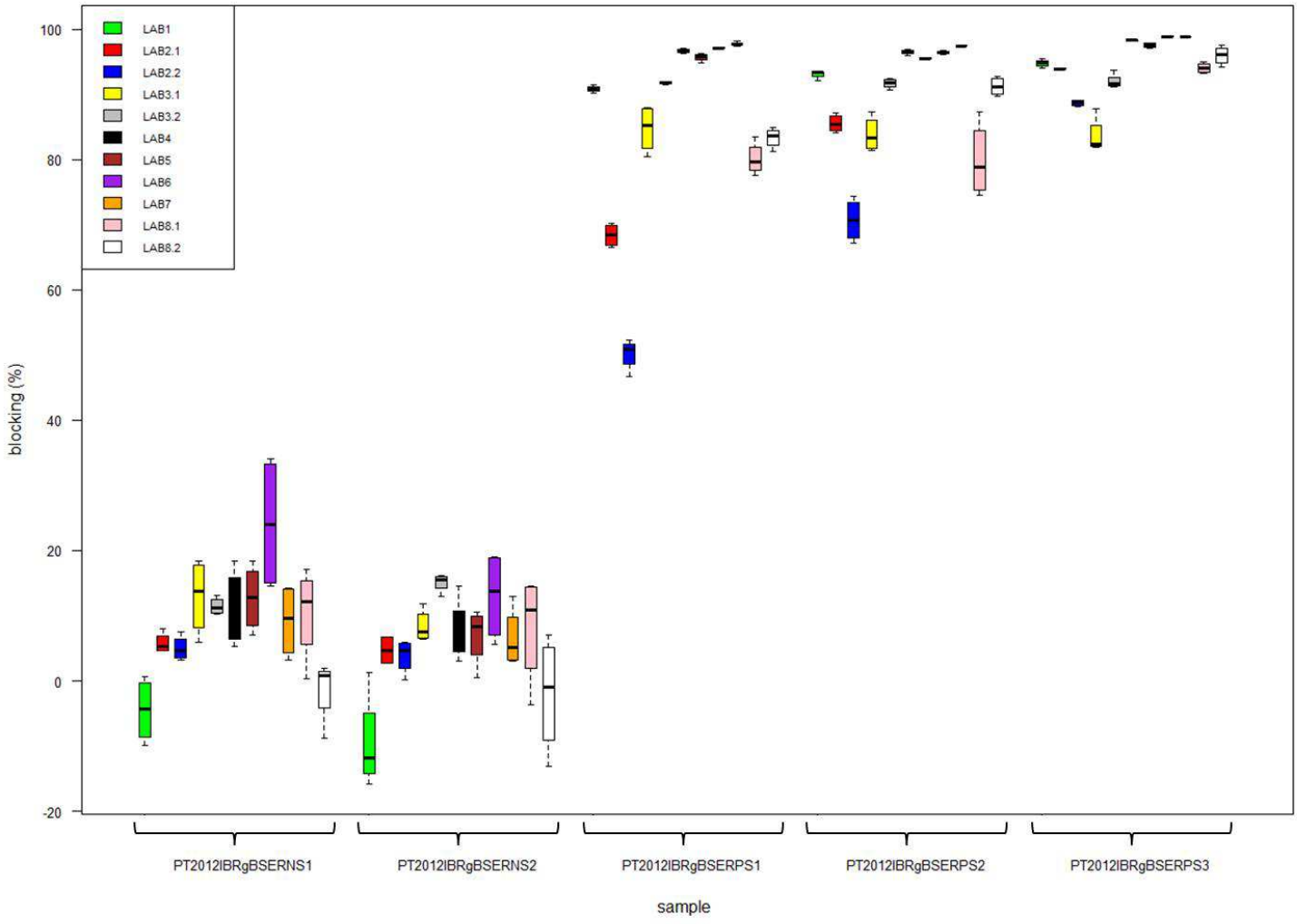

Figure 1. Box plots showing the percentage blocking per IBRgB reference serum sample and per participating (sub)laboratory. Box plots represent the minimum value, the maximum value, the median, the lower (25%) and upper (75%) quartile, and possible outliers per sample and per laboratory. Cut-off values are not shown since IBRgB ELISA kits from different producers and also different incubation protocols were used. LAB1, LAB3.2, LAB4, LAB5, LAB6 and LAB7 used an IBRgB ELISA kit from the same producer. LAB2.1 and LAB2.2 on the one hand, and LAB8.1 and LAB8.2 on the other hand, also used an IBRgB ELISA kit from the same producer.

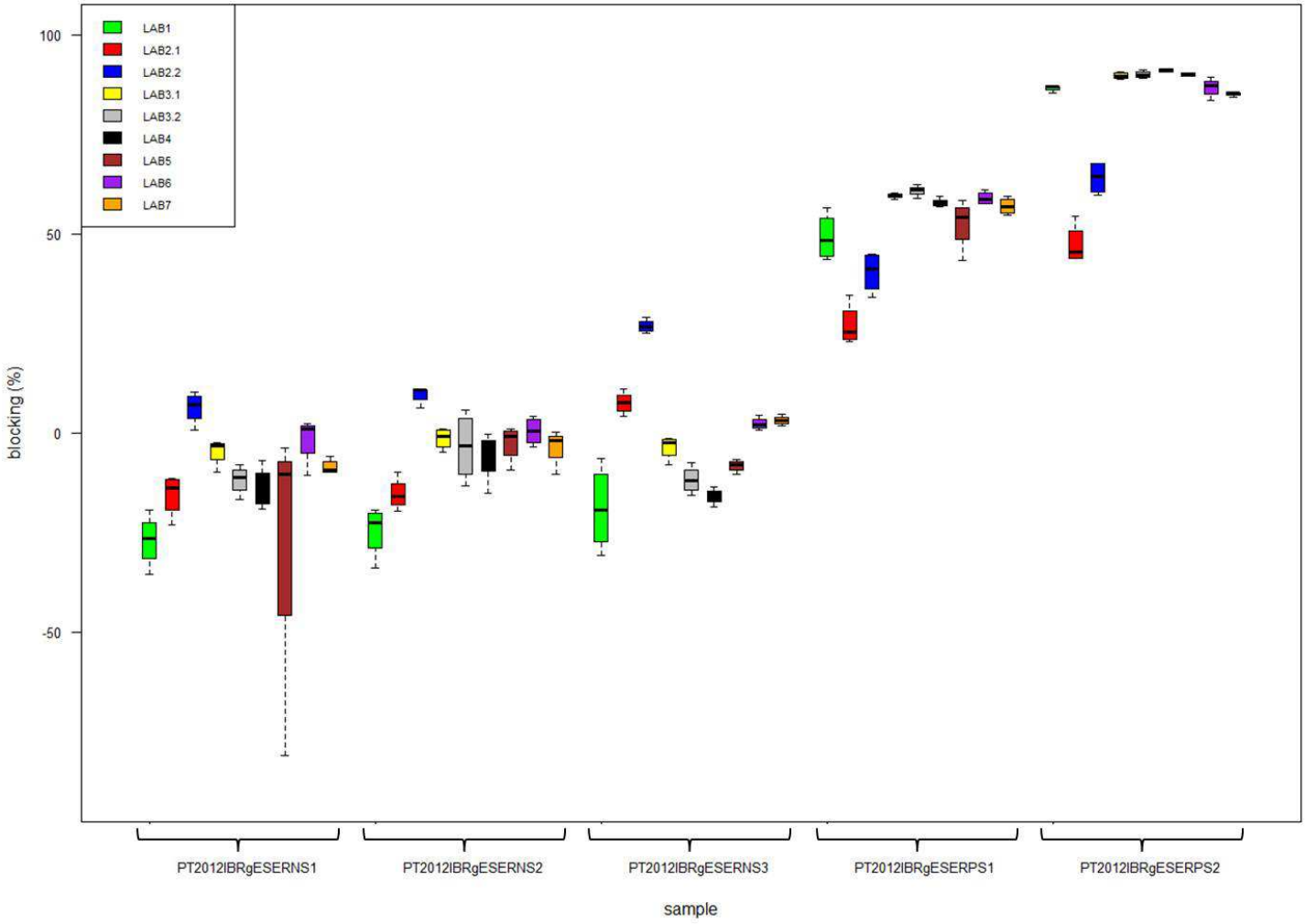

Figure 2. Box plots showing the percentage blocking per IBRgE reference serum sample and per participating (sub)laboratory. Box plots represent the minimum value, the maximum value, the median, the lower (25%) and upper (75%) quartile, and possible outliers per sample and per laboratory. Cut-off values are not shown since IBRgE ELISA kits from different producers and also different incubation protocols were used. LAB1, LAB3.1, LAB3.2, LAB4, LAB5, LAB6 and LAB7 on the one hand, and LAB2.1 and LAB2.2 on the other hand, used an IBRgE ELISA kit from the same producer.

II. Mandel's h- and k-statistics (z-scores)

Based on ISO 5725-2 and ISO 13528, between-lab variability (reproducibility) and within-lab variability (repeatability) were estimated through Mandel's h- and k-statistics, respectively, using the statistical software SAS 9.2. Mandel's h- and k-statistics were calculated based on the percentages blocking (calculated according to the instructions for this PT) per reference serum sample and per participating laboratory.

The h-statistic depends on the number of participants, whereas the k-statistic depends on both the number of participants and the number of repeats per sample. When 30 participants or more are involved in a PT, a satisfactory between-lab and within-lab consistency is obtained when the (absolute) value for the h- and k-statistic is smaller than 2. An unsatisfactory result (a corrective action is required) is reached when the (absolute) value is larger than 3. (Absolute) values between 2 and 3 indicate a questionable consistency. Importantly, in case of a smaller number of participants (which is the case in this PT), other indicator values apply for Mandel's h- and k-statistics (Table 1).

Table 1. Indicators for Mandel's h- and k-statistics at the 5% significance level in function of the number of participating laboratories (p) and the number of repeats per sample (n) as described in ISO 5725-2.

p (# labs)	h	k								
		n (# repeats)								
		2	3	4	5	6	7	8	9	10
3	1,15	1,65	1,53	1,45	1,40	1,37	1,34	1,32	1,30	1,29
4	1,42	1,76	1,59	1,50	1,44	1,40	1,37	1,35	1,33	1,31
5	1,57	1,81	1,62	1,53	1,46	1,42	1,39	1,36	1,34	1,32
6	1,66	1,85	1,64	1,54	1,48	1,43	1,40	1,37	1,35	1,33
7	1,71	1,87	1,66	1,55	1,49	1,44	1,41	1,38	1,36	1,34
8	1,75	1,88	1,67	1,56	1,50	1,45	1,41	1,38	1,36	1,34
9	1,78	1,90	1,68	1,57	1,50	1,45	1,42	1,39	1,36	1,35
10	1,80	1,90	1,68	1,57	1,50	1,46	1,42	1,39	1,37	1,35
11	1,82	1,91	1,69	1,58	1,51	1,46	1,42	1,39	1,37	1,35

Based on Table 1, the maximum absolute value for Mandel's h-statistic is 1,82 for the IBRgB PT (p=11) and 1,78 for the IBRgE PT (p=9). The maximum value for Mandel's k-statistic is 1,58 for the IBRgB PT (p=11 and n=4) and 1,57 for the IBRgE PT (p=9 and n=4).

For the detection of IBRgB-specific antibodies, 7 out of 11 participating (sub)laboratories obtained a satisfactory between-laboratory consistency for all reference serum samples: LAB2.1, LAB3.2, LAB4, LAB5, LAB7, LAB8.1 and LAB8.2. The other participants showed an increased value for Mandel's h-statistic for at least 1 reference serum sample: LAB1 for the negative reference serum sample PT2012IBRgBSERNS2 (h=-2,26), LAB2.2 for the positive reference serum samples PT2012IBRgBSERPS1 (h=-2,38) and PT2012IBRgBSERPS2 (h=-2,20), LAB3.1 for the positive reference serum sample PT2012IBRgBSERPS3 (h=-2,25), and LAB6 for the negative reference serum sample PT2012IBRgBSERNS1 (h=2,00). LAB1, LAB3.2, LAB4, LAB5, LAB6 and LAB7 used an IBRgB ELISA kit from the same producer. LAB2.1 and LAB2.2 on the one hand, and LAB8.1 and LAB8.2 on the other hand, also used an IBRgB ELISA kit from the same producer.
Furthermore, 7 out of 11 participating (sub)laboratories obtained a satisfactory within-laboratory consistency for all reference serum samples: LAB1, LAB2.1, LAB2.2, LAB3.2, LAB4, LAB5 and LAB7. The other participants showed an increased value for Mandel's k-statistic for at least 1 reference serum sample: LAB3.1 for the positive reference serum samples PT2012IBRgBSERPS1 (k=2,15) and PT2012IBRgBSERPS3 (k=2,61), LAB6 for the negative reference serum sample PT2012IBRgBSERNS1 (k=1,87), LAB8.1 for the positive reference serum sample PT2012IBRgBSERPS2 (k=2,55), and LAB8.2 for the negative reference serum sample PT2012IBRgBSERNS2 (k=1,62).

For the detection of IBRgE-specific antibodies, 6 out of 9 participating (sub)laboratories obtained a satisfactory between-laboratory consistency for all reference serum samples: LAB3.1, LAB3.2, LAB4, LAB5, LAB6 and LAB7. The other participants showed an increased value for Mandel's h-statistic for at least 1 reference serum sample: LAB1 for the negative reference serum sample PT2012IBRgESERNS2 (h=-1,99), LAB2.1 for the positive reference serum samples PT2012IBRgESERPS1 (h=-2,18) and PT2012IBRgESERPS2 (h=-2,23), and LAB2.2 for the negative reference serum sample PT2012IBRgESERNS3 (h=2,05). LAB1, LAB3.1, LAB3.2, LAB4, LAB5, LAB6 and LAB7 on the one hand, and LAB2.1 and LAB2.2 on the other hand, used an IBRgE ELISA kit from the same producer.
Only 4 out of 9 participating (sub)laboratories obtained a satisfactory within-laboratory consistency for all reference serum samples: LAB3.1, LAB4, LAB6 and LAB7. The other participants showed an increased value for Mandel's k-statistic for at least 1 reference serum sample: LAB1 for the negative reference serum sample PT2012IBRgESERNS3 (k=2,56), LAB2.1 for the positive reference serum sample PT2012IBRgESERPS2 (k=2,11), LAB2.2 for the positive reference serum sample PT2012IBRgESERPS2 (k=1,72), LAB3.2 for the negative reference serum sample PT2012IBRgESERNS2 (k=1,65), and LAB5 for the negative reference serum sample PT2012IBRgESERNS1 (k=2,82) and the positive reference serum sample PT2012IBRgESERPS1 (k=1,62).

All data used for the calculations of Mandel's h- and k-statistics can be found in Annex 2.

III. ANOVA

Using a SAS macro encoding a general linear model (GLM) with laboratories as fixed effect and the normalized OD values (in this case the percentages blocking calculated according to the instructions for this PT) as a dependent variable, it was investigated whether statistically significant differences exist (α=0,05) between participating laboratories and between different kits, batches or incubation protocols used at the same laboratory. Comparisons were made at the global level (all reference serum samples were analysed together), status level (all reference serum samples with the same status were analysed together) and sample level (all reference serum samples were analysed individually). Since comparing quantitative results between participants or methods (e.g. different kits, batches or incubation protocols) is most relevant at the status level (less variation than at a global level), we focused on the latter.

III.1. IBRgB

For the IBRgB PT, no statistically significant differences were observed between laboratories or between different kits, batches or incubation protocols used at the same laboratory at a global level. However, statistically significant differences existed at both sample and status level. At the status level, significant differences were observed for both the negative and positive reference serum samples.

The short and long incubation protocol of the same batch of the IBRgB ELISA kit used by LAB2 (LAB2.1 and LAB2.2, respectively) did result in significant differences for the positive but not for the negative reference serum samples. The percentages blocking for the positive reference serum samples obtained by LAB2.2 were significantly lower than those obtained by LAB2.1 and the other participants. In contrast, the short and long incubation protocol of the same batch of the IBRgB ELISA kit used by LAB8 (LAB8.2 and LAB8.1, respectively) did result in significant differences for the negative but not for the positive reference serum samples. The percentages blocking for the negative reference serum samples reported by LAB8.2 were significantly lower than those reported by LAB8.1. The percentages blocking obtained by LAB3 using IBRgB ELISA kits from 2 different producers (LAB3.1 and LAB3.2) were not significantly different, neither for the negative nor for the positive reference serum samples.

III.2. IBRgE

For the IBRgE PT, no statistically significant differences were observed between laboratories or between different kits, batches or incubation protocols used at the same laboratory at a global level. However, statistically significant differences existed at both sample and status level. At the status level, significant differences were observed for both the negative and positive reference serum samples.

The short and long incubation protocol of the same batch of the IBRgE ELISA kit used by LAB2 (LAB2.1 and LAB2.2, respectively) did result in significant differences for the negative but not for the positive reference serum samples. The percentages blocking for the negative reference serum samples obtained by LAB2.2 were significantly higher than those obtained by LAB2.1 and the other participants. In contrast, the percentages blocking obtained by LAB3 using 2 different batches of the same IBRgE ELISA kit (LAB3.1 and LAB3.2) were not significantly different, neither for the negative nor for the positive reference serum samples.

Annex 2: Calculations of Mandel's h- and k-statistics (based on % blocking)

A. IBRgB

Sample	Labnr	n_i	v_i	x_i_m	x_g_m	between_ lab_coeff	STDEV _repeat	STDEV _repro	STDEV _betweenlab	h	k	cv
PT2012IBRgBSERNS1	1	4	24,35	-4,44	8,83	0,14	5,64	6,07	2,25	-1,74	0,87	-111,11
PT2012IBRgBSERNS1	2.1	4	2,38	5,81	8,83	0,14	5,64	6,07	2,25	-0,40	0,27	26,59
PT2012IBRgBSERNS1	2.2	4	3,65	4,96	8,83	0,14	5,64	6,07	2,25	-0,51	0,34	38,55
PT2012IBRgBSERNS1	3.1	4	34,19	12,98	8,83	0,14	5,64	6,07	2,25	0,54	1,04	45,07
PT2012IBRgBSERNS1	3.2	4	1,76	11,45	8,83	0,14	5,64	6,07	2,25	0,34	0,23	11,57
PT2012IBRgBSERNS1	4	4	35,19	11,10	8,83	0,14	5,64	6,07	2,25	0,30	1,05	53,42
PT2012IBRgBSERNS1	5	4	26,21	12,71	8,83	0,14	5,64	6,07	2,25	0,51	0,91	40,30
PT2012IBRgBSERNS1	6	4	111,65	24,15	8,83	0,14	5,64	6,07	2,25	2,00	1,87	43,76
PT2012IBRgBSERNS1	7	4	33,12	9,23	8,83	0,14	5,64	6,07	2,25	0,05	1,02	62,36
PT2012IBRgBSERNS1	8.1	4	52,42	10,47	8,83	0,14	5,64	6,07	2,25	0,21	1,28	69,18
PT2012IBRgBSERNS1	8.2	4	24,86	-1,28	8,83	0,14	5,64	6,07	2,25	-1,32	0,88	-388,41
PT2012IBRgBSERNS2	1	4	56,12	-9,54	5,70	0,11	5,59	5,92	1,94	-2,26	1,34	-78,53
PT2012IBRgBSERNS2	2.1	4	5,44	4,71	5,70	0,11	5,59	5,92	1,94	-0,15	0,42	49,58
PT2012IBRgBSERNS2	2.2	4	7,32	3,85	5,70	0,11	5,59	5,92	1,94	-0,27	0,48	70,22
PT2012IBRgBSERNS2	3.1	4	6,36	8,35	5,70	0,11	5,59	5,92	1,94	0,39	0,45	30,18
PT2012IBRgBSERNS2	3.2	4	1,99	15,08	5,70	0,11	5,59	5,92	1,94	1,39	0,25	9,36
PT2012IBRgBSERNS2	4	4	24,01	7,61	5,70	0,11	5,59	5,92	1,94	0,28	0,88	64,39
PT2012IBRgBSERNS2	5	4	19,66	6,93	5,70	0,11	5,59	5,92	1,94	0,18	0,79	64,02
PT2012IBRgBSERNS2	6	4	48,13	12,99	5,70	0,11	5,59	5,92	1,94	1,08	1,24	53,41
PT2012IBRgBSERNS2	7	4	20,65	6,52	5,70	0,11	5,59	5,92	1,94	0,12	0,81	69,71
PT2012IBRgBSERNS2	8.1	4	72,94	8,20	5,70	0,11	5,59	5,92	1,94	0,37	1,53	104,17
PT2012IBRgBSERNS2	8.2	4	81,65	-1,99	5,70	0,11	5,59	5,92	1,94	-1,14	1,62	-453,97
PT2012IBRgBSERPS1	1	4	0,29	90,79	85,12	0,88	1,70	4,94	4,63	0,39	0,32	0,59
PT2012IBRgBSERPS1	2.1	4	3,11	68,36	85,12	0,88	1,70	4,94	4,63	-1,14	1,04	2,58
PT2012IBRgBSERPS1	2.2	4	5,77	50,15	85,12	0,88	1,70	4,94	4,63	-2,38	1,41	4,79
PT2012IBRgBSERPS1	3.1	4	13,37	84,69	85,12	0,88	1,70	4,94	4,63	-0,03	2,15	4,32
PT2012IBRgBSERPS1	3.2	4	0,05	91,73	85,12	0,88	1,70	4,94	4,63	0,45	0,13	0,24
PT2012IBRgBSERPS1	4	4	0,12	96,71	85,12	0,88	1,70	4,94	4,63	0,79	0,20	0,36

Sample	Labnr	n_i	v_i	x_i_m	x_g_m	between_ lab_coeff	STDEV _repeat	STDEV _repro	STDEV _betweenlab	h	k	cv
PT2012IBRgBSERPS1	5	4	0,37	95,65	85,12	0,88	1,70	4,94	4,63	0,72	0,36	0,64
PT2012IBRgBSERPS1	6	4	0,02	97,08	85,12	0,88	1,70	4,94	4,63	0,81	0,09	0,15
PT2012IBRgBSERPS1	7	4	0,10	97,71	85,12	0,88	1,70	4,94	4,63	0,86	0,18	0,32
PT2012IBRgBSERPS1	8.1	4	6,30	80,10	85,12	0,88	1,70	4,94	4,63	-0,34	1,47	3,13
PT2012IBRgBSERPS1	8.2	4	2,42	83,34	85,12	0,88	1,70	4,94	4,63	-0,12	0,91	1,87
PT2012IBRgBSERPS2	1	4	0,43	93,09	89,24	0,57	2,29	3,49	2,64	0,46	0,29	0,71
PT2012IBRgBSERPS2	2.1	4	1,85	85,51	89,24	0,57	2,29	3,49	2,64	-0,44	0,60	1,59
PT2012IBRgBSERPS2	**2.2**	4	11,09	70,72	89,24	0,57	2,29	3,49	2,64	**-2,20**	1,46	4,71
PT2012IBRgBSERPS2	3.1	4	7,40	83,82	89,24	0,57	2,29	3,49	2,64	-0,64	1,19	3,25
PT2012IBRgBSERPS2	3.2	4	0,53	91,67	89,24	0,57	2,29	3,49	2,64	0,29	0,32	0,79
PT2012IBRgBSERPS2	4	4	0,13	96,47	89,24	0,57	2,29	3,49	2,64	0,86	0,16	0,38
PT2012IBRgBSERPS2	5	4	0,02	95,51	89,24	0,57	2,29	3,49	2,64	0,74	0,06	0,15
PT2012IBRgBSERPS2	6	4	0,07	96,42	89,24	0,57	2,29	3,49	2,64	0,85	0,12	0,28
PT2012IBRgBSERPS2	7	4	0,02	97,38	89,24	0,57	2,29	3,49	2,64	0,97	0,06	0,14
PT2012IBRgBSERPS2	**8.1**	4	33,91	79,86	89,24	0,57	2,29	3,49	2,64	-1,12	**2,55**	7,29
PT2012IBRgBSERPS2	8.2	4	2,05	91,20	89,24	0,57	2,29	3,49	2,64	0,23	0,63	1,57
PT2012IBRgBSERPS3	1	4	0,29	94,74	94,21	0,66	1,06	1,82	1,48	0,11	0,50	0,57
PT2012IBRgBSERPS3	2.1	4	0,01	93,87	94,21	0,66	1,06	1,82	1,48	-0,07	0,10	0,11
PT2012IBRgBSERPS3	2.2	4	0,23	88,67	94,21	0,66	1,06	1,82	1,48	-1,18	0,45	0,54
PT2012IBRgBSERPS3	**3.1**	4	7,73	83,59	94,21	0,66	1,06	1,82	1,48	**-2,25**	**2,61**	3,33
PT2012IBRgBSERPS3	3.2	4	1,32	92,00	94,21	0,66	1,06	1,82	1,48	-0,47	1,08	1,25
PT2012IBRgBSERPS3	4	4	0,01	98,31	94,21	0,66	1,06	1,82	1,48	0,87	0,11	0,12
PT2012IBRgBSERPS3	5	4	0,14	97,45	94,21	0,66	1,06	1,82	1,48	0,69	0,35	0,38
PT2012IBRgBSERPS3	6	4	0,01	98,85	94,21	0,66	1,06	1,82	1,48	0,98	0,10	0,11
PT2012IBRgBSERPS3	7	4	0,01	98,83	94,21	0,66	1,06	1,82	1,48	0,98	0,09	0,09
PT2012IBRgBSERPS3	8.1	4	0,66	94,06	94,21	0,66	1,06	1,82	1,48	-0,03	0,76	0,87
PT2012IBRgBSERPS3	8.2	4	2,06	95,96	94,21	0,66	1,06	1,82	1,48	0,37	1,35	1,49

Legend: **Labnr** = number attributed to a laboratory during the PT; **n_i** = number of replicates; **v_i** = total variability (variance) in the normalised data (% blocking); **x_i_m** = mean of normalized data (% blocking); **x_g_m** = mean of normalized data (% blocking) obtained by all laboratories; b**etween_lab_coeff** = fraction of total variability due to differences between labs for each sample; **STDEV_repeat** = repeatability standard deviation over all laboratories; **STDEV_repro** = reproducibility standard deviation over all laboratories; **STDEV_betweenlab** = between-lab standard deviation over all laboratories; **h-statistic** = between-laboratory consistency; **k-statistic** = within-laboratory consistency; **CV** = variation coefficient in %. Values for Mandel's h- and k-statistics shown in red/underlined/bold exceed the corresponding limit value as determined in Annex 1 (Table 1).

B. IBRgE

Sample	Labnr	n_i	v_i	x_i_m	x_g_m	between_ lab_coeff	STDEV _repeat	STDEV _repro	STDEV _betweenlab	h	k	cv
PT2012IBRgESERNS1	1	4	44,01	-27,02	-11,46	0,05	13,00	13,37	3,11	-1,42	0,51	-24,56
PT2012IBRgESERNS1	2.1	4	28,43	-15,49	-11,46	0,05	13,00	13,37	3,11	-0,37	0,41	-34,42
PT2012IBRgESERNS1	2.2	4	16,26	6,37	-11,46	0,05	13,00	13,37	3,11	1,63	0,31	63,30
PT2012IBRgESERNS1	3.1	4	11,41	-4,71	-11,46	0,05	13,00	13,37	3,11	0,62	0,26	-71,76
PT2012IBRgESERNS1	3.2	4	13,87	-11,81	-11,46	0,05	13,00	13,37	3,11	-0,03	0,29	-31,54
PT2012IBRgESERNS1	4	4	26,93	-13,85	-11,46	0,05	13,00	13,37	3,11	-0,22	0,40	-37,48
PT2012IBRgESERNS1	5	4	1340,33	-26,42	-11,46	0,05	13,00	13,37	3,11	-1,37	2,82	-138,60
PT2012IBRgESERNS1	6	4	36,21	-1,63	-11,46	0,05	13,00	13,37	3,11	0,90	0,46	-368,75
PT2012IBRgESERNS1	7	4	3,87	-8,60	-11,46	0,05	13,00	13,37	3,11	0,26	0,15	-22,87
PT2012IBRgESERNS2	1	4	42,27	-24,60	-5,16	0,29	5,18	6,15	3,32	-1,99	1,26	-26,43
PT2012IBRgESERNS2	2.1	4	16,03	-15,31	-5,16	0,29	5,18	6,15	3,32	-1,04	0,77	-26,14
PT2012IBRgESERNS2	2.2	4	4,98	9,71	-5,16	0,29	5,18	6,15	3,32	1,53	0,43	22,98
PT2012IBRgESERNS2	3.1	4	6,67	-1,35	-5,16	0,29	5,18	6,15	3,32	0,39	0,50	-191,66
PT2012IBRgESERNS2	3.2	4	73,42	-3,47	-5,16	0,29	5,18	6,15	3,32	0,17	1,65	-246,92
PT2012IBRgESERNS2	4	4	41,64	-5,68	-5,16	0,29	5,18	6,15	3,32	-0,05	1,25	-113,59
PT2012IBRgESERNS2	5	4	22,17	-2,59	-5,16	0,29	5,18	6,15	3,32	0,26	0,91	-181,50
PT2012IBRgESERNS2	6	4	12,79	0,38	-5,16	0,29	5,18	6,15	3,32	0,57	0,69	945,36
PT2012IBRgESERNS2	7	4	21,33	-3,52	-5,16	0,29	5,18	6,15	3,32	0,17	0,89	-131,23
PT2012IBRgESERNS3	1	4	113,38	-18,95	-2,10	0,58	4,17	6,45	4,93	-1,20	2,56	-56,18
PT2012IBRgESERNS3	2.1	4	8,04	7,51	-2,10	0,58	4,17	6,45	4,93	0,68	0,68	37,77
PT2012IBRgESERNS3	2.2	4	2,96	26,82	-2,10	0,58	4,17	6,45	4,93	2,05	0,41	6,41
PT2012IBRgESERNS3	3.1	4	9,13	-3,62	-2,10	0,58	4,17	6,45	4,93	-0,11	0,72	-83,51
PT2012IBRgESERNS3	3.2	4	12,07	-11,79	-2,10	0,58	4,17	6,45	4,93	-0,69	0,83	-29,48
PT2012IBRgESERNS3	4	4	4,22	-16,04	-2,10	0,58	4,17	6,45	4,93	-0,99	0,49	-12,80
PT2012IBRgESERNS3	5	4	2,60	-8,25	-2,10	0,58	4,17	6,45	4,93	-0,44	0,39	-19,52
PT2012IBRgESERNS3	6	4	2,38	2,29	-2,10	0,58	4,17	6,45	4,93	0,31	0,37	67,29
PT2012IBRgESERNS3	7	4	1,49	3,15	-2,10	0,58	4,17	6,45	4,93	0,37	0,29	38,76
PT2012IBRgESERPS1	1	4	35,56	49,19	51,49	0,49	3,98	5,57	3,89	-0,21	1,50	12,12
PT2012IBRgESERPS1	2.1	4	27,67	27,10	51,49	0,49	3,98	5,57	3,89	-2,18	1,32	19,41
PT2012IBRgESERPS1	2.2	4	26,50	40,39	51,49	0,49	3,98	5,57	3,89	-0,99	1,29	12,74

Sample	Labnr	n_i	v_i	x_i_m	x_g_m	between_ lab_coeff	STDEV _repeat	STDEV _repro	STDEV _betweenlab	h	k	cv
PT2012IBRgESERPS1	3.1	4	0,39	59,53	51,49	0,49	3,98	5,57	3,89	0,72	0,16	1,05
PT2012IBRgESERPS1	3.2	4	2,06	60,79	51,49	0,49	3,98	5,57	3,89	0,83	0,36	2,36
PT2012IBRgESERPS1	4	4	1,48	57,79	51,49	0,49	3,98	5,57	3,89	0,56	0,31	2,10
PT2012IBRgESERPS1	**5**	4	41,76	52,59	51,49	0,49	3,98	5,57	3,89	0,10	**1,62**	12,29
PT2012IBRgESERPS1	6	4	2,46	58,96	51,49	0,49	3,98	5,57	3,89	0,67	0,39	2,66
PT2012IBRgESERPS1	7	4	4,57	57,07	51,49	0,49	3,98	5,57	3,89	0,50	0,54	3,75
PT2012IBRgESERPS2	1	4	0,65	86,69	81,24	0,83	2,38	5,86	5,35	0,36	0,34	0,93
PT2012IBRgESERPS2	**2.1**	4	25,26	47,34	81,24	0,83	2,38	5,86	5,35	**-2,23**	**2,11**	10,62
PT2012IBRgESERPS2	**2.2**	4	16,85	64,13	81,24	0,83	2,38	5,86	5,35	-1,13	**1,72**	6,40
PT2012IBRgESERPS2	3.1	4	0,63	89,78	81,24	0,83	2,38	5,86	5,35	0,56	0,33	0,88
PT2012IBRgESERPS2	3.2	4	0,70	90,03	81,24	0,83	2,38	5,86	5,35	0,58	0,35	0,93
PT2012IBRgESERPS2	4	4	0,17	91,10	81,24	0,83	2,38	5,86	5,35	0,65	0,17	0,45
PT2012IBRgESERPS2	5	4	0,30	90,09	81,24	0,83	2,38	5,86	5,35	0,58	0,23	0,60
PT2012IBRgESERPS2	6	4	6,14	86,83	81,24	0,83	2,38	5,86	5,35	0,37	1,04	2,85
PT2012IBRgESERPS2	7	4	0,33	85,19	81,24	0,83	2,38	5,86	5,35	0,26	0,24	0,67

Legend: **Labnr** = number attributed to a laboratory during the PT; **n_i** = number of replicates; **v_i** = total variability (variance) in the normalised data (% blocking); **x_i_m** = mean of normalized data (% blocking); **x_g_m** = mean of normalized data (% blocking) obtained by all laboratories; b**etween_lab_coeff** = fraction of total variability due to differences between labs for each sample; **STDEV_repeat** = repeatability standard deviation over all laboratories; **STDEV_repro** = reproducibility standard deviation over all laboratories; **STDEV_betweenlab** = between-lab standard deviation over all laboratories; **h-statistic** = between-laboratory consistency; **k-statistic** = within-laboratory consistency; **CV** = variation coefficient in %. Values for Mandel's h- and k-statistics shown in red/underlined/bold exceed the corresponding limit value as determined in Annex 1 (Table 1).