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172-PT

PROFICIENCY TESTING 2016

Q-FEVER (QFV)

***Detection of QFV-specific antibodies in serum and/or milk by
Enzyme Linked Immunosorbent Assay (ELISA)***

CODA-CERVA-UCCLE

DATE BEGIN PT: 23 MAY 2016

DATE REPORT: 23 AUGUST 2016

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 QFV-specific antibodies in serum and/or milk samples of bovidae origin by ELISA.

III. Materials and methods

III.1. Conduct of diagnostic tests

In the framework of this PT, predefined reference serum and/or milk samples must be tested by means of a QFV antibody 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. Reference serum samples

Replicates of 5 reference serum samples of bovine origin, either free from detectable QFV-specific antibodies (n=2; coded 'PT2016QFVSERNS1' and 'PT2016QFVSERNS2') or containing detectable QFV-specific antibodies (n=3; coded 'PT2016QFVSERPS1', 'PT2016QFVSERPS2' and 'PT2016QFVSERPS3'), were used. In total, 120 aliquots were distributed to 6 participating laboratories. All participants received 4 aliquots of each reference serum sample, i.e. 20 aliquots in total. The identification numbers of the reference serum samples were randomized for all participants (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 PT2016QFVSERNS2, PT2016QFVSERPS1, and PT2016QFVSERPS2 was based on the results obtained during pre-verification, hereby using the LSIVET ruminant milk/serum QFV Antibody ELISA Test Kit from LSI, the CHEKIT QFV Antibody ELISA Test Kit from IDEXX and the ID Screen[®] QFV Indirect Multi-species Antibody ELISA Test Kit from IDVET. For these reference serum samples, the same qualitative result was obtained with all ELISA kits used. The status of the reference serum samples PT2016QFVSERNS1 and PT2016QFVSERPS3 was based on the results obtained using the LSIVET ruminant milk/serum QFV Antibody ELISA Test Kit from LSI. Taken together, the reference serum samples PT2016QFVSERNS1 and PT2016QFVSERNS2 were considered as negative samples, the reference serum samples PT2016QFVSERPS1, PT2016QFVSERPS2, and PT2016QFVSERPS3 as positive samples.

A homogeneity check on the aliquoted reference serum samples had been performed as in the context of PTs under the procedure PRO/2.5/01. Indeed, 10 aliquots of each reference serum sample were analysed using the LSIVET ruminant milk/serum QFV Antibody ELISA Test Kit from LSI, 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 the participating laboratories to correctly identify the absence or presence of QFV-specific antibodies in 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 LSIVET ruminant milk/serum QFV Antibody ELISA Test Kit from LSI.

III.2.2. Reference milk samples

Replicates of 5 reference milk samples of bovidae origin, either free from detectable QFV-specific antibodies (n=2; coded 'PT2016QFVSERNM1' and 'PT2016QFVSERNM2') or containing detectable QFV-specific antibodies (n=3; coded 'PT2016QFVSERPM1', 'PT2016QFVSERPM2' and 'PT2016QFVSERPM3'), were used. The reference milk samples PT2016QFVSERNM1, PT2016QFVSERNM2, PT2016QFVSERPM1 and PT2016QFVSERPM2 were bovine tank milk, whereas the reference milk sample PT2016QFVSERPM3 was caprine tank milk.

In total, 40 aliquots were distributed to the 2 participating laboratories. All participants received 4 aliquots of each reference milk sample, i.e. 20 aliquots in total. The identification numbers of the reference milk samples were randomized for all participants (Table 5).

For each reference milk sample, a certificate containing the status of the sample (= 'golden standard') was made. The status of the reference milk samples was based on the results obtained during pre-verification, hereby using the LSIVET ruminant milk/serum QFV Antibody ELISA Test Kit from LSI, the CHEKIT QFV Antibody ELISA Test Kit from IDEXX and the ID Screen[®] QFV Indirect Multi-species Antibody ELISA Test Kit from IDVET. For all reference milk samples, the same qualitative result was obtained with all ELISA kits used. Taken together, the reference milk samples PT2016QFVSERNM1 and PT2016QFVSERNM2 were considered as negative samples and the reference milk samples PT2016QFVSERPM1, PT2016QFVSERPM2 and PT2016QFVSERPM3 as positive samples in QFV antibody ELISA.

A homogeneity check on the aliquoted reference milk samples had already been performed in the context of previous PTs Serology QFV. Indeed, 10 aliquots of each reference milk sample were analysed using the LSIVET ruminant milk/serum QFV Antibody ELISA Test Kit from LSI, hereby obtaining the same qualitative result for all 10 aliquots of the same reference milk sample. Consequently, all reference milk samples were considered as reliable samples in order to evaluate the ability of the participating laboratories to correctly identify the absence or presence of QFV-specific antibodies in milk. In addition, all reference milk samples were tested once after the PT in order to confirm their stability and status (post-verification) using the LSIVET ruminant milk/serum QFV Antibody ELISA Test Kit from LSI.

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* when the reported result matches with the assigned status or *failure* when the reported result does not match with the assigned status.

III.3.2. Level of agreement

The level of agreement achieved by the participating laboratories is expressed as the percentage of *success* for the 20 aliquots of reference samples used for either 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 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 CODA-CERVA-Uccle.

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

LAB1, LAB2 participated in both the PT serum and the PT milk and hence received 40 aliquots: 20 aliquots of reference serum samples and 20 aliquots of reference milk samples. In contrast, LAB3, LAB4, LAB5 and LAB6 only participated in the PT serum and hence received 20 aliquots of reference serum samples.

Lyophilized reference serum samples (120 aliquots in total) and lyophilized reference milk samples (40 aliquots in total) were sent at ambient temperature to the 6 participating laboratories by national or international courier on the 23th of May 2016. LAB1, LAB2 and LAB4 acknowledged receipt of the samples on 23th of May 2016, whereas the other participants acknowledged receipt of the samples on 25th of May 2016. Analyses were performed between 24th of May 2016 and 7th of June 2016 (Table 1).

IV.2. Dates at which results were returned to the CODA-CERVA-Uccle

Results were submitted to the CODA-CERVA-Uccle between 3th and 7th of June 2016 (Table 1). All participants hereby respected the deadline of 10th of June 2016 for submission of the results.

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

Laboratory	Reference samples received	Start of analysis SERUM	Start of analysis MILK	Submission of the results (Excel file)
LAB1	23/05/2016	31/05/2016	31/05/2016	03/06/2016
LAB2	23/05/2016	07/06/2016	24/05/2016	07/06/2016
LAB3	25/05/2016	25/05/2016	NA	06/06/2016
LAB4	23/05/2016	24/05/2016	NA	06/06/2016
LAB5	25/05/2016	01/06/2016	NA	06/06/2016
LAB6	25/05/2016	31/05/2016	NA	07/06/2016

Legend: NA = not applicable

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

IV.4.1. Level of agreement

Qualitative data analysis showed that:

- (i) For the detection of QFV-specific antibodies in **serum**, all 6 participating laboratories provided qualitative results that were in full agreement with the assigned status of the reference serum samples and thus achieved 100% of agreement (Table 2).
- (ii) For the detection of QFV-specific antibodies in **milk**, the 2 participating laboratories provided qualitative results that were in full agreement with the assigned status of the reference milk samples and thus achieved 100% of agreement (Table 3).

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

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

	LABNR					
	1	2	3	4	5	6
failure	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)

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

	LABNR	
	1	2
failure	0 (0.0)	0 (0.0)
success	20 (100.0)	20 (100.0)

IV.4.2. Variability among participating laboratories

Variability in the qualitative laboratory results of the participating laboratories was not observed with any of the reference serum and milk samples.

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

Table 4. The responses (RESULT) of the participating laboratories (LABNR) with the internal identification of the reference serum samples (SAMPLE), the external identification of the reference serum samples (LABPOSIT), and the status assigned by the QFV reference laboratory of CODA-CERVA-Uccle (STATUS). NEG: negative; POS: positive.

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
1	1	1	PT2016QFVSERPS1	POS	POS	1
2	1	2	PT2016QFVSERNS1	NEG	NEG	1
3	1	3	PT2016QFVSERPS2	POS	POS	1
4	1	4	PT2016QFVSERPS2	POS	POS	1
5	1	5	PT2016QFVSERPS3	POS	POS	1
6	1	6	PT2016QFVSERNS2	NEG	NEG	1
7	1	7	PT2016QFVSERPS2	POS	POS	1
8	1	8	PT2016QFVSERPS1	POS	POS	1
9	1	9	PT2016QFVSERNS2	NEG	NEG	1
10	1	10	PT2016QFVSERNS1	NEG	NEG	1
11	1	11	PT2016QFVSERPS3	POS	POS	1
12	1	12	PT2016QFVSERPS3	POS	POS	1
13	1	13	PT2016QFVSERNS2	NEG	NEG	1
14	1	14	PT2016QFVSERPS3	POS	POS	1
15	1	15	PT2016QFVSERNS1	NEG	NEG	1
16	1	16	PT2016QFVSERPS1	POS	POS	1
17	1	17	PT2016QFVSERPS2	POS	POS	1
18	1	18	PT2016QFVSERNS1	NEG	NEG	1
19	1	19	PT2016QFVSERNS2	NEG	NEG	1
20	1	20	PT2016QFVSERPS1	POS	POS	1
21	2	1	PT2016QFVSERPS2	POS	POS	1
22	2	2	PT2016QFVSERNS2	NEG	NEG	1
23	2	3	PT2016QFVSERNS1	NEG	NEG	1
24	2	4	PT2016QFVSERPS3	POS	POS	1
25	2	5	PT2016QFVSERPS1	POS	POS	1
26	2	6	PT2016QFVSERNS2	NEG	NEG	1
27	2	7	PT2016QFVSERNS1	NEG	NEG	1
28	2	8	PT2016QFVSERPS3	POS	POS	1
29	2	9	PT2016QFVSERPS2	POS	POS	1
30	2	10	PT2016QFVSERNS2	NEG	NEG	1
31	2	11	PT2016QFVSERPS1	POS	POS	1
32	2	12	PT2016QFVSERNS1	NEG	NEG	1
33	2	13	PT2016QFVSERPS3	POS	POS	1
34	2	14	PT2016QFVSERPS2	POS	POS	1
35	2	15	PT2016QFVSERPS3	POS	POS	1
36	2	16	PT2016QFVSERPS1	POS	POS	1
37	2	17	PT2016QFVSERNS2	NEG	NEG	1
38	2	18	PT2016QFVSERPS1	POS	POS	1
39	2	19	PT2016QFVSERPS2	POS	POS	1
40	2	20	PT2016QFVSERNS1	NEG	NEG	1



(Table 4 - CONTINUED)

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
41	3	1	PT2016QFVSERPS1	POS	POS	1
42	3	2	PT2016QFVSERNS1	NEG	NEG	1
43	3	3	PT2016QFVSERPS2	POS	POS	1
44	3	4	PT2016QFVSERPS2	POS	POS	1
45	3	5	PT2016QFVSERPS3	POS	POS	1
46	3	6	PT2016QFVSERNS2	NEG	NEG	1
47	3	7	PT2016QFVSERPS2	POS	POS	1
48	3	8	PT2016QFVSERPS1	POS	POS	1
49	3	9	PT2016QFVSERNS2	NEG	NEG	1
50	3	10	PT2016QFVSERNS1	NEG	NEG	1
51	3	11	PT2016QFVSERPS3	POS	POS	1
52	3	12	PT2016QFVSERPS3	POS	POS	1
53	3	13	PT2016QFVSERNS2	NEG	NEG	1
54	3	14	PT2016QFVSERPS3	POS	POS	1
55	3	15	PT2016QFVSERNS1	NEG	NEG	1
56	3	16	PT2016QFVSERPS1	POS	POS	1
57	3	17	PT2016QFVSERPS2	POS	POS	1
58	3	18	PT2016QFVSERNS1	NEG	NEG	1
59	3	19	PT2016QFVSERNS2	NEG	NEG	1
60	3	20	PT2016QFVSERPS1	POS	POS	1
61	4	1	PT2016QFVSERPS2	POS	POS	1
62	4	2	PT2016QFVSERNS2	NEG	NEG	1
63	4	3	PT2016QFVSERNS1	NEG	NEG	1
64	4	4	PT2016QFVSERPS3	POS	POS	1
65	4	5	PT2016QFVSERPS1	POS	POS	1
66	4	6	PT2016QFVSERNS2	NEG	NEG	1
67	4	7	PT2016QFVSERNS1	NEG	NEG	1
68	4	8	PT2016QFVSERPS3	POS	POS	1
69	4	9	PT2016QFVSERPS2	POS	POS	1
70	4	10	PT2016QFVSERNS2	NEG	NEG	1
71	4	11	PT2016QFVSERPS1	POS	POS	1
72	4	12	PT2016QFVSERNS1	NEG	NEG	1
73	4	13	PT2016QFVSERPS3	POS	POS	1
74	4	14	PT2016QFVSERPS2	POS	POS	1
75	4	15	PT2016QFVSERPS3	POS	POS	1
76	4	16	PT2016QFVSERPS1	POS	POS	1
77	4	17	PT2016QFVSERNS2	NEG	NEG	1
78	4	18	PT2016QFVSERPS1	POS	POS	1
79	4	19	PT2016QFVSERPS2	POS	POS	1
80	4	20	PT2016QFVSERNS1	NEG	NEG	1



(Table 4 - CONTINUED)

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
81	5	1	PT2016QFVSERPS1	POS	POS	1
82	5	2	PT2016QFVSERNS1	NEG	NEG	1
83	5	3	PT2016QFVSERPS2	POS	POS	1
84	5	4	PT2016QFVSERPS2	POS	POS	1
85	5	5	PT2016QFVSERPS3	POS	POS	1
86	5	6	PT2016QFVSERNS2	NEG	NEG	1
87	5	7	PT2016QFVSERPS2	POS	POS	1
88	5	8	PT2016QFVSERPS1	POS	POS	1
89	5	9	PT2016QFVSERNS2	NEG	NEG	1
90	5	10	PT2016QFVSERNS1	NEG	NEG	1
91	5	11	PT2016QFVSERPS3	POS	POS	1
92	5	12	PT2016QFVSERPS3	POS	POS	1
93	5	13	PT2016QFVSERNS2	NEG	NEG	1
94	5	14	PT2016QFVSERPS3	POS	POS	1
95	5	15	PT2016QFVSERNS1	NEG	NEG	1
96	5	16	PT2016QFVSERPS1	POS	POS	1
97	5	17	PT2016QFVSERPS2	POS	POS	1
98	5	18	PT2016QFVSERNS1	NEG	NEG	1
99	5	19	PT2016QFVSERNS2	NEG	NEG	1
100	5	20	PT2016QFVSERPS1	POS	POS	1
101	6	1	PT2016QFVSERPS2	POS	POS	1
102	6	2	PT2016QFVSERNS2	NEG	NEG	1
103	6	3	PT2016QFVSERNS1	NEG	NEG	1
104	6	4	PT2016QFVSERPS3	POS	POS	1
105	6	5	PT2016QFVSERPS1	POS	POS	1
106	6	6	PT2016QFVSERNS2	NEG	NEG	1
107	6	7	PT2016QFVSERNS1	NEG	NEG	1
108	6	8	PT2016QFVSERPS3	POS	POS	1
109	6	9	PT2016QFVSERPS2	POS	POS	1
110	6	10	PT2016QFVSERNS2	NEG	NEG	1
111	6	11	PT2016QFVSERPS1	POS	POS	1
112	6	12	PT2016QFVSERNS1	NEG	NEG	1
113	6	13	PT2016QFVSERPS3	POS	POS	1
114	6	14	PT2016QFVSERPS2	POS	POS	1
115	6	15	PT2016QFVSERPS3	POS	POS	1
116	6	16	PT2016QFVSERPS1	POS	POS	1
117	6	17	PT2016QFVSERNS2	NEG	NEG	1
118	6	18	PT2016QFVSERPS1	POS	POS	1
119	6	19	PT2016QFVSERPS2	POS	POS	1
120	6	20	PT2016QFVSERNS1	NEG	NEG	1



Table 5. The responses (RESULT) of the participating laboratories (LABNR) with the internal identification of the reference milk samples (SAMPLE), the external identification of the reference milk samples (LABPOSIT), and the status assigned by the QFV reference laboratory of CODA-CERVA-Uccle (STATUS). NEG: negative; POS: positive.

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
1	1	1	PT2016QFVSERNM1	NEG	NEG	1
2	1	2	PT2016QFVSERNM2	NEG	NEG	1
3	1	3	PT2016QFVSERPM1	POS	POS	1
4	1	4	PT2016QFVSERPM2	POS	POS	1
5	1	5	PT2016QFVSERPM3	POS	POS	1
6	1	6	PT2016QFVSERNM2	NEG	NEG	1
7	1	7	PT2016QFVSERPM2	POS	POS	1
8	1	8	PT2016QFVSERNM1	NEG	NEG	1
9	1	9	PT2016QFVSERPM3	POS	POS	1
10	1	10	PT2016QFVSERPM3	POS	POS	1
11	1	11	PT2016QFVSERNM2	NEG	NEG	1
12	1	12	PT2016QFVSERPM2	POS	POS	1
13	1	13	PT2016QFVSERPM3	POS	POS	1
14	1	14	PT2016QFVSERPM1	POS	POS	1
15	1	15	PT2016QFVSERNM1	NEG	NEG	1
16	1	16	PT2016QFVSERNM2	NEG	NEG	1
17	1	17	PT2016QFVSERPM2	POS	POS	1
18	1	18	PT2016QFVSERPM1	POS	POS	1
19	1	19	PT2016QFVSERPM1	POS	POS	1
20	1	20	PT2016QFVSERNM1	NEG	NEG	1
21	2	1	PT2016QFVSERPM1	POS	POS	1
22	2	2	PT2016QFVSERNM1	NEG	NEG	1
23	2	3	PT2016QFVSERPM3	POS	POS	1
24	2	4	PT2016QFVSERNM1	NEG	NEG	1
25	2	5	PT2016QFVSERPM2	POS	POS	1
26	2	6	PT2016QFVSERPM3	POS	POS	1
27	2	7	PT2016QFVSERNM2	NEG	NEG	1
28	2	8	PT2016QFVSERPM1	POS	POS	1
29	2	9	PT2016QFVSERPM2	POS	POS	1
30	2	10	PT2016QFVSERNM2	NEG	NEG	1
31	2	11	PT2016QFVSERPM3	POS	POS	1
32	2	12	PT2016QFVSERNM1	NEG	NEG	1
33	2	13	PT2016QFVSERNM2	NEG	NEG	1
34	2	14	PT2016QFVSERPM1	POS	POS	1
35	2	15	PT2016QFVSERPM3	POS	POS	1
36	2	16	PT2016QFVSERPM2	POS	POS	1
37	2	17	PT2016QFVSERNM2	NEG	NEG	1
38	2	18	PT2016QFVSERNM1	NEG	NEG	1
39	2	19	PT2016QFVSERPM2	POS	POS	1
40	2	20	PT2016QFVSERPM1	POS	POS	1

V. Discussion

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

For the detection of QFV-specific antibodies in serum, all participating laboratories provided qualitative results that were in full agreement with the assigned status of the reference serum samples (100% of agreement) (Table 2 and Table 4).

QFV antibody ELISA kits were from 2 different producers and different batches from the same ELISA kit were used: IDEXX (1 batch: E121) and LSI (3 batches: 2ELISACOXLS-025, 2ELISACOXLS-027, 5ELISACOXLS-026). LAB1, LAB2, LAB4, LAB5 and LAB6 used a QFV antibody ELISA kit from the same producer. In addition, LAB1 and LAB4 on the one hand, and LAB5 and LAB6 on the other hand used the same batch.

For the detection of QFV-specific antibodies in milk, all participating laboratories provided qualitative results that were in full agreement with the assigned status of the reference milk samples (100% of agreement) (Table 3 and Table 5).

The participating laboratories used QFV antibody ELISA kits from the same producer but different batches: LSI (batches: 2ELISACOXLS-025, 2ELISACOXLS-027).

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 samples assigned by CODA-CERVA-Uccle (see III.3.3.). Consequently, all participants in the PT serum achieved a satisfactory performance for the detection of QFV-specific antibodies in reference serum samples and all participants in the PT milk achieved a satisfactory performance for the detection of QFV-specific antibodies in reference milk samples.

Coordinator proficiency tests

Katia Knapen

Appendix

Name of the participating laboratories

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

Animal and Plant Health Agency (APHA) (Weybridge, United-Kingdom)

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

Dierengezondheidszorg Vlaanderen (DGZ) (Torhout, Belgium)

LNCR / ACSEDIATE (Maisons-Alfort, France)

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

Annex 1: Quantitative data analysis (Box plots)

Besides qualitative data analysis (positive, negative or non-interpretable result), also quantitative data analysis was performed using the statistical software programs R (box plots).

Box plots represent the minimum and maximum value that are not considered as outliers, the 25th and 75th percentile (respectively P25 and P75), the median (P50), and possible outliers per sample and per laboratory. Values lower than $(P25 - 1.5(P75 - P25))$ and higher than $(P75 + 1.5(P75 - P25))$ are considered as outliers. Note that due to the low number of data available, outliers cannot be detected when the number of data is smaller than 5 and $P25 = \text{minimum}$ and $P75 = \text{maximum}$ when the number data is 2.

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.

For the **antibody ELISA serum reference samples**, box plots of the normalized OD values according the PT provider per reference sample and per participating laboratory are shown in Figure 1.

Figure 1 (antigen ELISA serum reference samples)

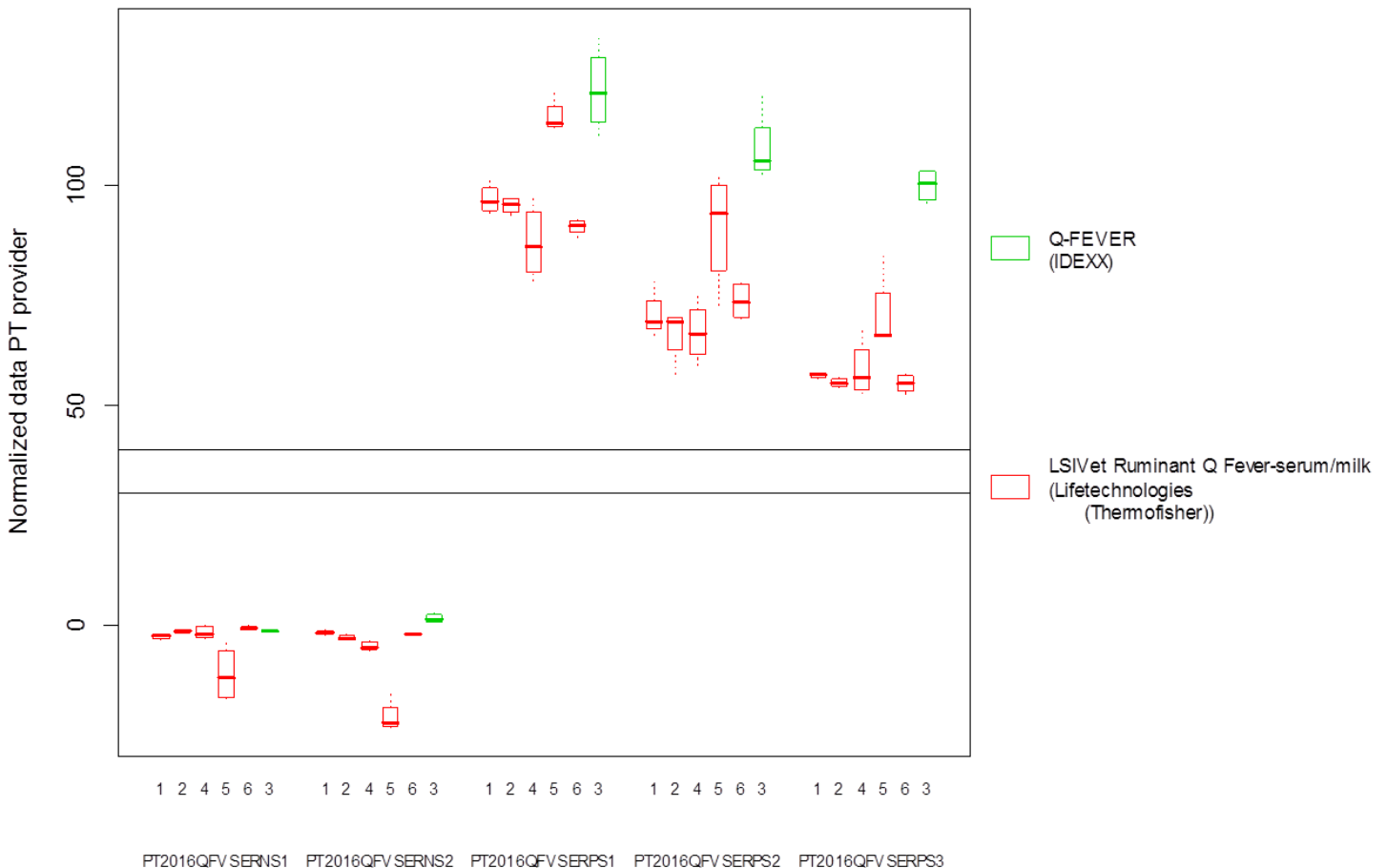


Figure 1. Box plots showing the normalized OD values according the PT provider per reference serum and per participating laboratory. LAB1, LAB2, LAB4, LAB5 and LAB6 used a QFV antibody ELISA kit from the same producer. In addition, LAB1 and LAB4 on the one hand, and LAB5 and LAB6 on the other hand used the same batch. Cut-off values (IDEXX 30 and LSI 40) are shown by horizontal lines.

For the **antibody ELISA milk reference samples**, box plots of the normalized OD values according the PT provider per reference sample and per participating laboratory are shown in Figure 2.

Figure 2 (antigen ELISA milk reference samples)

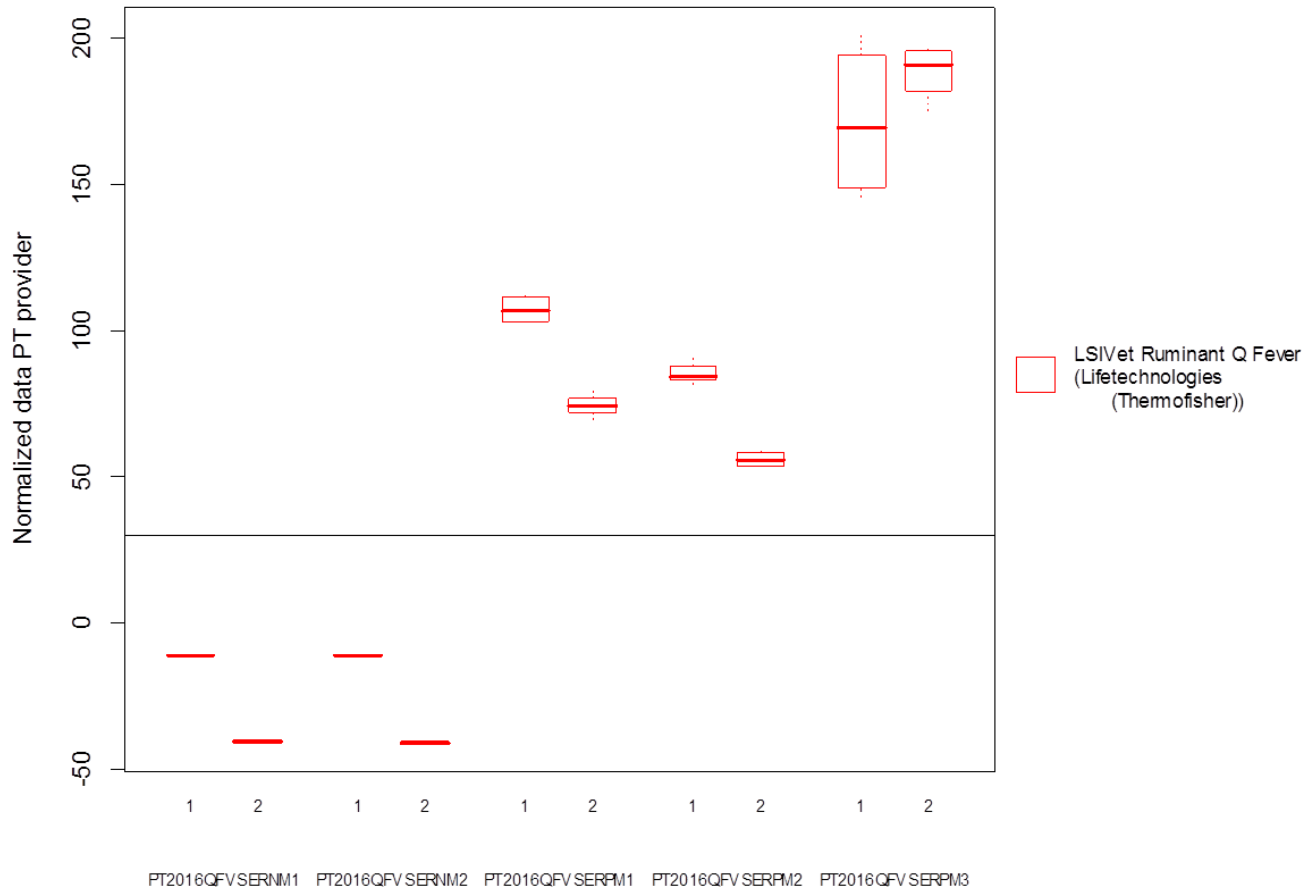


Figure 1. Box plots showing the normalized OD values according the PT provider per reference milk and per participating laboratory. The participating laboratories used QFV antibody ELISA kits from the same producer but different batches. Cut-off value is shown by a horizontal line.