



CODA-CERVA

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172-TEST

PROFICIENCY TESTING 2011

Scrapie (SCR)

***Genotype identification for the detection
of genetically linked susceptibility to scrapie in blood***

**OPERATIONAL UNIT
COORDINATION OF VETERINARY DIAGNOSIS
EPIDEMIOLOGY AND RISK ASSESSMENT
(CVD-ERA)**

DATE BEGIN PT: 21 MARCH 2011

DATE REPORT: 18 APRIL 2011

I. Introduction

Details relevant to the proficiency test are available in the procedure PRO/2.5/01 'Beheer van de proficiency testen/Gestion des essais d'aptitude'.

II. Aim

This proficiency test focused on genotype identification for the detection of genetically linked susceptibility to scrapie (SCR) in blood of sheep origin and aimed to assess the analytical accuracy of the tests conducted by the participants.

III. Materials and methods

III.1. Conduct of diagnostic tests

In the framework of this proficiency test, predefined reference blood samples must be tested by means of RT-PCR. The procedures for this RT-PCR must be fully described in the SOPs of the participating laboratories.

III.2. Reference samples

Ten reference blood samples of sheep origin, coded 'PT2011SCRGENB1', 'PT2011SCRGENB2', 'PT2011SCRGENB3', 'PT2011SCRGENB4', 'PT2011SCRGENB5', 'PT2011SCRGENB6', 'PT2011SCRGENB7', 'PT2011SCRGENB8', 'PT2011SCRGENB9' and 'PT2011SCRGENB10', were used. In total, 40 aliquots, prepared by the reference laboratory for SCR of the Veterinary and Agrochemical Research Center (CODA-CERVA), were distributed to the participating laboratories. Each participant was given one aliquot of each reference blood sample. The positions of the reference blood samples in the sent blocks were randomized for each participant (Table 2).

For the reference blood samples, a certificate containing the assigned genotypes was made by the reference laboratory for SCR of CODA-CERVA (status of the sample = 'golden standard'). The assigned genotypes were obtained by testing each reference blood sample once before the proficiency test (pre-verification) by three different assays (RT-PCR, DGGE-RFLP, sequencing), hereby obtaining each time the same result. Consequently, these reference blood samples were considered as reliable samples to use for the purpose of this proficiency test. In addition, the reference blood samples were also tested once after the proficiency test in order to confirm their stability and status (post-verification).

III.3. Qualitative data analysis

III.3.1. Classification of results

Results provided by the participating laboratories are categorized as *success* (if the genotype was correctly identified) or *failure* (if the genotype was not correctly identified).

III.3.2. Level of agreement

The level of agreement achieved by a participating laboratory is expressed as the percentage *success* for all ten reference blood samples used in this proficiency test.

III.3.3. Threshold for qualification

Following the procedure, a participating laboratory is only qualified if at least 90% of the reference blood samples are analysed correctly, i.e. when the reported result corresponds with the status assigned by the reference laboratory for SCR of CODA-CERVA.

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 the CODA-CERVA.

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

The ten reference blood samples were sent on 21st of March 2011 to each of the four participating laboratories (40 aliquots in total). All laboratories acknowledged receipt of the samples on the same day. The analyses were carried out on 22nd (LAB4), 24th (LAB2) and 31st (LAB1) of March 2011, and on 1st of April 2011 (LAB3).

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

Results from the participating laboratories have been received on 31st of March 2011 (LAB2), 1st of April 2011 (LAB1 and LAB4), and 4th of April 2011 (LAB3). LAB3 hereby exceeded the deadline of 1st of April 2011 for the delivery of the results.

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

LAB2, LAB3 and LAB4 reached 100% of agreement for genotype identification for the detection of genetically linked susceptibility to scrapie in reference blood samples. LAB1 identified only 80% of the genotypes correctly (Table 1).

Table 1. Agreement between results obtained by the participating laboratories (LABNR) and the status of the reference blood samples assigned by the reference laboratory for SCR of CODA-CERVA. All participating laboratories received 10 reference blood samples. Results are presented as absolute values and percentages (in parentheses).

	LABNR			
	1	2	3	4
failure	2 (20.0)	0 (0.0)	0 (0.0)	0 (0.0)
success	8 (80.0)	10 (100.0)	10 (100.0)	10 (100.0)

IV.4.2 Variability among participating laboratories

No variability between LAB2, LAB3 and LAB4 could be observed since these participants identified all reference blood samples correctly. LAB1 misidentified sample PT2011SCRGENB6 (ALRR/ALRQ instead of ALRR/VLRQ) and could not report a result for sample PT2011SCRGENB7. For each participating laboratory, the obtained responses for the reference blood samples are shown in Table 2.



Table 2. The responses (RESULT) of the participating laboratories (LABNR) with the identification of the reference blood samples (SAMPLE), the position of the reference blood samples as placed in the block (LABPOSIT), and the status assigned by the reference laboratory for SCR of the CODA-CERVA (STATUS).

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
1	1	1	PT2011SCRGENB1	ALRR/VLRQ	ALRR/VLRQ	1
2	1	2	PT2011SCRGENB2	ALRR/ALRH	ALRR/ALRH	1
3	1	3	PT2011SCRGENB3	ALRR/ALRR	ALRR/ALRR	1
4	1	4	PT2011SCRGENB4	ALRR/ALRQ	ALRR/ALRQ	1
5	1	5	PT2011SCRGENB5	ALRQ/ALRH	ALRH/ALRQ	1
6	1	6	PT2011SCRGENB6	<u>ALRR/VLRQ</u>	<u>ALRR/ALRQ</u>	<u>0</u>
7	1	7	PT2011SCRGENB7	<u>ALRQ/ALHQ</u>	<u>séquence non lisible</u>	<u>0</u>
8	1	8	PT2011SCRGENB8	ALRQ/ALRQ	ALRQ/ALRQ	1
9	1	9	PT2011SCRGENB9	ALRR/ALRQ	ALRR/ALRQ	1
10	1	10	PT2011SCRGENB10	VLRQ/VLRQ	VLRQ/VLRQ	1
11	2	1	PT2011SCRGENB3	ALRR/ALRR	ALRR/ALRR	1
12	2	2	PT2011SCRGENB4	ALRR/ALRQ	ALRR / ALRQ	1
13	2	3	PT2011SCRGENB5	ALRQ/ALRH	ALRQ / ALRH	1
14	2	4	PT2011SCRGENB6	ALRR/VLRQ	ALRR / VLRQ	1
15	2	5	PT2011SCRGENB7	ALRQ/ALHQ	ALRQ / ALHQ	1
16	2	6	PT2011SCRGENB8	ALRQ/ALRQ	ALRQ / ALRQ	1
17	2	7	PT2011SCRGENB9	ALRR/ALRQ	ALRR / ALRQ	1
18	2	8	PT2011SCRGENB10	VLRQ/VLRQ	VLRQ / VLRQ	1
19	2	9	PT2011SCRGENB1	ALRR/VLRQ	ALRR / VLRQ	1
20	2	10	PT2011SCRGENB2	ALRR/ALRH	ALRR / ALRH	1
21	3	1	PT2011SCRGENB5	ALRQ/ALRH	ALRQ/ALRH	1
22	3	2	PT2011SCRGENB6	ALRR/VLRQ	ALRR/VLRQ	1
23	3	3	PT2011SCRGENB7	ALRQ/ALHQ	ALRQ/ALHQ	1
24	3	4	PT2011SCRGENB8	ALRQ/ALRQ	ALRQ/ALRQ	1
25	3	5	PT2011SCRGENB9	ALRR/ALRQ	ALRR/ALRQ	1
26	3	6	PT2011SCRGENB10	VLRQ/VLRQ	VLRQ/VLRQ	1
27	3	7	PT2011SCRGENB1	ALRR/VLRQ	ALRR/VLRQ	1
28	3	8	PT2011SCRGENB2	ALRR/ALRH	ALRR/ALRH	1
29	3	9	PT2011SCRGENB3	ALRR/ALRR	ALRR/ALRR	1
30	3	10	PT2011SCRGENB4	ALRR/ALRQ	ALRR/ALRQ	1

(Table 2 - CONTINUED)

	LABNR	LABPOSIT	SAMPLE	STATUS	RESULT	SUCCESS
31	4	1	PT2011SCRGENB7	ALRQ/ALHQ	ALRQ/ALHQ	1
32	4	2	PT2011SCRGENB8	ALRQ/ALRQ	ALRQ/ALRQ	1
33	4	3	PT2011SCRGENB9	ALRR/ALRQ	ALRR/ALRQ	1
34	4	4	PT2011SCRGENB10	VLRQ/VLRQ	VLRQ/VLRQ	1
35	4	5	PT2011SCRGENB1	ALRR/VLRQ	ALRR/VLRQ	1
36	4	6	PT2011SCRGENB2	ALRR/ALRH	ALRR/ALRH	1
37	4	7	PT2011SCRGENB3	ALRR/ALRR	ALRR/ALRR	1
38	4	8	PT2011SCRGENB4	ALRR/ALRQ	ALRR/ALRQ	1
39	4	9	PT2011SCRGENB5	ALRQ/ALRH	ALRQ/ALRH	1
40	4	10	PT2011SCRGENB6	ALRR/VLRQ	ALRR/VLRQ	1

V. Discussion

The purpose of this proficiency test was to assess the performance of the participating laboratories when analyzing reference blood samples from sheep by PCR in order to identify genotypes genetically linked to susceptibility to scrapie.

Data obtained in this proficiency test showed that three out of four participating laboratories provided responses that were in full agreement with the true status of the reference blood samples. One participating laboratory (LAB1) reached 80% of agreement with the true status of the reference blood samples. This participant reported for sample PT2011SCRGENB6 a different genotype than the assigned genotype, whereas no result could be reported for sample PT2011SCRGENB7.

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 blood samples assigned by the reference laboratory for SCR of the CODA-CERVA (see III.3.3.). Consequently, LAB2, LAB3 and LAB4 achieved a satisfactory performance since they provided qualitative results that were in full agreement with the true status of the reference blood samples. In contrast, LAB1 obtained only 80% of agreement with the true status of the reference blood samples and therefore did not achieve a satisfactory performance.

Head CVD-ERA
Yves Van der Stede

Appendix

Name of the participating Laboratories

ARSIA (Mons)

CODA-CERVA

Progenus

Quality Partner