

**BIOLOGICAL HEALTH RISKS  
QUALITY OF LABORATORIES**

**COMMITTEE OF EXPERTS**

**EXTERNAL QUALITY ASSESSMENT  
IN VETERINARY DIAGNOSIS**

**DEFINITIVE GLOBAL ANNUAL REPORT**

**VETERINARY MEDECINE**

**2022**

**Sciensano/PT VET/3-E**

Biological health risks  
Quality of laboratories  
J. Wytsmanstreet, 14  
1050 Brussels | Belgium

[www.sciensano.be](http://www.sciensano.be)

**COMMITTEE OF EXPERTS  
NATIONAL REFERENCE LABORATORIES**

<b>Sciensano</b>			
Secretariat		PHONE: 02/642.55.22	FAX: 02/642.56.45
Ynse Van de Maele	Scheme coordinator	PHONE: 02/642 55 24	
		e-mail: Ynse.VandeMaele@sciensano.be	
Bernard China	Alternate coordinator	PHONE: 02/642 53 85	
		e-mail: Bernard.China@sciensano.be	
<b>Experts</b>	<b>Institute</b>		
Marylene Tignon	<b>Sciensano - Enzootic, vector-borne and bee diseases</b>		
Gaëtan De Gryse	<b>Sciensano - Enzootic, vector-borne and bee diseases</b>		
Sylvie Marché	<b>Sciensano- Veterinary bacteriology</b>		
Cécile Boland	<b>Sciensano- Veterinary bacteriology</b>		
Marcella Mori	<b>Sciensano - Veterinary bacteriology</b>		
Anneleen Matthijs	<b>Sciensano- Veterinary bacteriology</b>		

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**Authorization of the report:** by Ynse Van de Maele, scheme coordinator

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- NL: <https://www.sciensano.be/nl/kwaliteit-van-laboratoria>
- FR: <https://www.sciensano.be/fr/qualite-des-laboratoires>
- EN: <https://www.sciensano.be/en/quality-laboratories>

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# 1 INTRODUCTION

## 1.1 Abbreviations

Below you can find a list of abbreviations (Table I) used throughout the global annual report.

**Table I:** List of abbreviations.

Abbreviation	Full name
Ab	Antibody
Ag	Antigen
ASF	African Swine Fever (type II strain)
AUJ	Aujeszky's disease
BRU	Brucellosis
BT	Bovine Tuberculosis
BVD	Bovine Viral Diarrhea
CAPX	Capripox
CSF	Classical Swine Fever
EBL	Enzootic Bovine Leukosis
EDTA	Ethylenediamine tetraacetic acid
ELISA	Enzyme-linked immunoassay
FASFC	Federal Agency for the Safety of the Food Chain
IBR	Infectious Bovine Rhinotracheitis
IFN $\gamma$	Interferon gamma
N	Number of participants
ND	Not determined
NR	Number of results
NCR	Number of correct results
PRRS	Porcine Reproductive and Respiratory Syndrome
PT	Proficiency test
QF	Q-Fever
RT-qPCR	Quantitative reverse transcription Polymer Chain Reaction
SAL	Salmonella pullorum-gallinarum
VM	Visna Maedi

## 1.2 Calendar

In 2022, 14 PTs were organized by Sciensano for the proficiency testing in the diagnosis of pathogens in veterinary medicine (Table II) following the ISO17043:2010 standard.

**Table II.** Proficiency tests (PTs) organized in 2022.

Name of proficiency test			Concerned methods	Send in the week of	Deadline
PRRS	Serology	Serum	ELISA (Ab)	28 February	28 March
	Virology*	Blood	RT-qPCR		
QF	Serology	Serum	ELISA (Ab)	14 March	8 April
	Serology	Milk	ELISA (Ab)		

Name of proficiency test			Concerned methods	Send in the week of	Deadline
ASF	Serology	Serum	ELISA (Ab)	21 March	15 April
	Virology	Serum	ELISA (Ab)		
CSF	Serology	Serum	ELISA (Ab)	28 March	22 April
CAPX**	Serology	Serum	Different methods	2 May	9 June
	Virology	Virus stock and tissue suspension	Different methods		
IBR	Serology	Serum gB	ELISA (Ab)	16 May	3 June
	Serology	Serum gE	ELISA (Ab)		
AUJ	Serology	Serum gB	ELISA (Ab)	6 June	1 July
	Serology	Serum gE	ELISA (Ab)		
EBL	Serology	Serum	ELISA (Ab)	20 June	15 July
BRU	Serology	Milk	ELISA (Ab)	26 September	21 October
	Bacteriology	Organs	Isolation		
BVD	Serology	Serum	ELISA (Ab)	10 October	4 November
	Serology***	Milk	ELISA (Ab)		
	Virology	Serum	ELISA (Ag)		
	Virology	EDTA-blood	ELISA (Ag)		
	Virology	Ear notch	ELISA (Ag)		
	Virology	Serum	RT-qPCR		
Virology	EDTA-blood	RT-qPCR			
Virology	Ear notch	RT-qPCR			
BT	Serology	Serum	ELISA (Ab)	24 October	18 November
	Gamma interferon	Serum	ELISA (Ab)		
LEPT	Bacteriology	Organs	Isolation	7 November	2 December
VM	Serology	Serum	ELISA (Ab)	29 November	16 December
SAL	Bacteriology	Organs	Isolation	5 December	23 December

\* = The PT of PRRS part virology on blood could not be organized this year due to staff shortages (covid crisis). This part will again be organized in 2023.

\*\* = The PT of CAPX is organized by the community reference laboratory, financed by the European Union and destined to the European reference laboratories. The results were not included in this global annual report.

\*\*\* = The PT of BVD part serology on milk was not and will not be organized in the future because there are very few laboratories in Europe that perform this analysis and therefore it was decided not to organize this PT anymore.

The calendar 2022 can be found on our website via these links:

- EN: <https://www.sciensano.be/en/biblio/eqa-calendar-2022>
- NL: <https://www.sciensano.be/nl/biblio/eke-kalender-2022>
- FR: <https://www.sciensano.be/nl/biblio/calendrier-eeq-2022>

## 1.3 Participants

Below you can find a list (Table III) of the number of participating laboratories in 2022. A distinction was made between accredited and non-accredited FASFC laboratories.

**Table III.** List of the number of participating laboratories in 2022.

Name of proficiency test			Concerned methods	FASFC	Other	Total
<b>PRRS</b>	Serology	Serum	ELISA (Ab)	4	3	7
<b>QF</b>	Serology	Serum	ELISA (Ab)	3	2	5
	Serology	Milk	ELISA (Ab)	2	1	3
<b>ASF</b>	Serology	Serum	ELISA (Ab)	3	3	6
	Virology	Serum	ELISA (Ab)	3	3	6
<b>CSF</b>	Serology	Serum	ELISA (Ab)	3	2	5
<b>IBR</b>	Serology	Serum gB	ELISA (Ab)	5	4	9
	Serology	Serum gE	ELISA (Ab)	5	6	11
<b>AUJ</b>	Serology	Serum gB	ELISA (Ab)	3	3	6
	Serology	Serum gE	ELISA (Ab)	4	6	10
<b>EBL</b>	Serology	Serum	ELISA (Ab)	3	3	6
<b>BRU</b>	Serology	Milk	ELISA (Ab)	4	1	5
	Bacteriology	Organs	Isolation	4	0	4
<b>BVD</b>	Serology	Serum	ELISA (Ab)	5	3	8
	Virology	Serum	ELISA (Ag)	5	1	6
	Virology	EDTA-blood	ELISA (Ag)	4	1	5
	Virology	Ear notch	ELISA (Ag)	5	1	6
	Virology	Serum	RT-qPCR	5	2	7
	Virology	EDTA-blood	RT-qPCR	3	3	6
	Virology	Ear notch	RT-qPCR	4	3	7
<b>BT</b>	Serology	Serum	ELISA (Ab)	4	1	5
	Gamma interferon	Serum	ELISA (Ab)	4	0	4
<b>LEPT</b>	Bacteriology	Organs	Isolation	3	0	3
<b>VM</b>	Serology	Serum	ELISA (Ab)	4	0	4
<b>SAL</b>	Bacteriology	Organs	Isolation	3	0	3

## 1.4 Criteria

The minimal required criteria (Table IV) for the qualification of a laboratory participating to the proficiency tests in veterinary medicine organized by Sciensano.

*Table IV: Criteria of acceptance.*

Test	Criteria for qualification
Tests with $\leq 5$ samples	Qualitative result (positive, negative, doubtful): <b>100%</b> of agreement between the results of the participating laboratory and the qualitative value (status) of the samples.
Tests with $> 5$ samples	Qualitative result (positive, negative, doubtful; genotype): <b><math>\geq 90\%</math></b> of agreement between the results of the participating laboratory and the qualitative value (status) of the samples.
Tests with $> 5$ samples	Strong positive samples: no mistakes allowed ( <b>100%</b> of agreement). Negative samples: 1 mistake allowed; Weak positive samples: 1 mistake allowed

## 1.5 Reports

The preliminary- and global report were placed on our webpage and can be find via these links:

- EN: <https://www.sciensano.be/en/external-quality-assessment/animal-health-pt-vet>
- NL: <https://www.sciensano.be/nl/externe-kwaliteitsevaluatie/diergezondheid-pt-vet>
- FR: <https://www.sciensano.be/fr/evaluation-externe-de-la-qualite/sante-animale-pt-vet>

## 2 RESULTS

### 2.1 Virology

The samples of this section were produced by the Enzootic, vector-borne and bee diseases laboratory of the directorate infectious diseases in animals of Sciensano.

#### 2.1.1 PORCINE REPRODUCTIVE AND RESPIRATORY SYNDROME (PRRS)

##### 2.1.1.1 Serology on serum

The panel consisted of 16 different samples, but samples NS1 and NS2 were repeated twice. Therefore, in total, the panel consisted of 18 samples (12 positive and 6 negative samples).

Two labs have chosen to test two different methods on the same samples, implying that there were two datasets submitted. These additional results are included in the tables below.

##### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS	1 (7)	7 POS
PS2	POS	1 (7)	7 POS
PS3	POS	1 (7)	7 POS
PS4	POS	1 (7)	7 POS
PS5	POS	1 (7)	7 POS
PS6	POS	1 (7)	7 POS
PS7	POS	1 (7)	7 POS
PS8	POS	1 (7)	7 POS
PS9	POS	1 (7)	7 POS
PS10	POS	1 (7)	7 POS
PS11	POS	1 (7)	7 POS
PS12	POS	1 (7)	7 POS
NS1	NEG	2 (14)	14 POS
NS2	NEG	2 (14)	14 POS
NS3	NEG	1 (7)	7 POS
NS4	NEG	1 (7)	7 POS

(POS = positive; NEG = negative)

##### Used method

Method	N	NR	NCR	%
IDEXX PRRS X3 Ab	5	90	90	100
Indical [Qiagen] – pigtype PRRSV Ab	1	18	18	100
BIOCHECK – PRRS XR	1	18	18	100
<b>TOTAL</b>	<b>7</b>	<b>126</b>	<b>126</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)



## Conclusion

In total, three different methods were used by the laboratories. All these methods achieved 100% correctness, which means that 126 correct results were submitted. As this was the first time a PT for PRRS serology on serum was organised, a high score was obtained for all laboratories.

## 2.1.2 AFRICAN SWINE FEVER (ASF)

### 2.1.2.1 Serology on serum

The panel consisted of 6 different samples, but samples PS2, PS3, PS4 and NS2 were repeated twice. Therefore, in total, the panel consisted of 10 samples (7 positive and 3 negative samples).

One lab had chosen to test two different methods on the same samples, implying that there were two datasets submitted. These additional results are included in the tables below.

#### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS	1 (6)	6 POS
PS2	POS	2 (12)	12 POS
PS3	POS	2 (12)	12 POS
PS4	POS	2 (12)	12 POS
NS1	NEG	1 (6)	6 NEG
NS2	NEG	2 (12)	12 NEG

(POS = positive; NEG = negative)

#### Used method

Method	N	NR	NCR	%
Ingenasa - Ingezym ASF-R	1	10	10	100
ID.VET - ID SCREEN® AFRICAN SWINE FEVER COMPETITION	4	40	40	100
Other	1	10	10	100
<b>TOTAL</b>	<b>6</b>	<b>60</b>	<b>60</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

## Conclusion

In total, three different methods were used by the laboratories. All these methods achieved 100% correctness, which means that 100 correct results were submitted.

### 2.1.2.2 Virology on serum

The panel consisted of 7 different samples, but samples PS4, PS5 and NS1 were repeated twice. Therefore, in total, the panel consisted of 10 samples (7 positive and 3 negative samples).

#### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS	1 (6)	6 POS
PS2	POS	1 (6)	6 POS
PS3	POS	1 (6)	6 POS
PS4	POS	2 (12)	12 POS
PS5	POS	2 (12)	12 POS
NS1	NEG	2 (12)	6 NEG
NS2	NEG	1 (6)	6 NEG

(POS = positive; NEG = negative)

#### Used method

Method	N	NR	NCR	%
PCR method: Tignon <i>et al</i> 2011	1	10	10	100
ID.VET - ID Gene® African Swine Fever Duplex	2	20	20	100
QIAGEN Virotype ASF PCR kit	1	10	10	100
Thermofisher - VetMAX™ African Swine Fever Virus Detection Kit	1	10	10	100
Idexx - RealPCR ASFV DNA mix lot	1	10	10	100
<b>TOTAL</b>	<b>6</b>	<b>60</b>	<b>60</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

#### Extraction method

Extraction method	N	NR	NCR	%
Indical - IndiMag Pathogen Kit	3	30	30	100
IDVET - ID Gene Mag Universal Extraction kit	1	10	10	100
ThermoFisher Scientific - other	1	10	10	100
QIAGEN - QIAamp DNA Mini kit	1	10	10	100
<b>TOTAL</b>	<b>6</b>	<b>60</b>	<b>60</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

#### Conclusion

In total, five different methods were used by the laboratories. All these methods achieved 100% correctness, which means that 100 correct results were submitted. One lab mentioned that their sample 1 and 4 only contained 250 µL each instead of 500 µL, but they reported that it was enough to perform the assay.

## 2.1.3 CLASSICAL SWINE FEVER (CSF)

### 2.1.3.1 Serology on serum

The panel consisted of 9 different samples. Samples PS1 and PS5 were repeated three times, whereas the other samples were repeated twice. Therefore, in total, the panel consisted of 20 samples (14 positive and 6 negative samples).

#### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS (weak)	3	15 POS
PS2	POS	2	10 POS
PS3	POS	2	10 POS
PS4	POS	2	10 POS
PS5	POS	3	15 POS
PS6	POS	2	10 POS
NS1	NEG	2	10 NEG
NS2	NEG	2	10 NEG
NS3	NEG	2	10 NEG

(POS = positive; NEG = negative)

#### Used method

Method	N	NR	NCR	%
Idexx - IDEXX CSFV Ab Test	5	100	100	100
<b>TOTAL</b>	<b>5</b>	<b>100</b>	<b>100</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

#### Conclusion

Only one method was used by the laboratories. This method achieved 100% correctness, which means that 100 correct results were submitted.

## 2.1.4 INFECTIOUS BOVINE RHINOTRACHEITIS (IBR)

### 2.1.4.1 Serology on serum gB

The panel consisted of 7 different samples. Samples PS1, PS3 and PS4 were repeated twice. Therefore, in total, the panel consisted of 10 samples (7 positive and 3 negative samples).

### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS	2 (18)	18 POS
PS2	POS	1 (9)	9 POS
PS3	POS – NEG – NI	2 (18)	13 NEG 5 NI
PS4	POS	2 (18)	18 POS
NS1	NEG	1 (9)	9 NEG
NS2	NEG	1 (9)	9 NEG
NS3	NEG	1 (9)	9 NEG

(POS = positive; NEG = negative)

### Used method

Method	N	NR	NCR	%
Idexx - IBR gB X3 Ab	9	90	90	100
<b>TOTAL</b>	<b>9</b>	<b>90</b>	<b>90</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

### Conclusion

Only one method was used by the laboratories. This method achieved 100% correctness, which means that 90 correct results were submitted.

#### 2.1.4.2 Serology on serum gE

The panel consisted of 6 different samples. Samples PS1, PS2, NS1 and NS3 were repeated twice. Therefore, in total, the panel consisted of 10 samples (5 positive and 5 negative samples).

One lab had chosen to test two different methods on the same samples, implying that there were two datasets submitted. These additional results are included in the tables below.

### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS	2 (22)	21 POS 1 NEG
PS2	POS	2 (22)	22 POS
PS3	POS	1 (11)	11 POS
NS1	NEG	2 (22)	22 NEG
NS2	NEG	1 (11)	11 NEG
NS3	NEG	2 (22)	22 NEG

(POS = positive; NEG = negative)

### Used method

Method	N	NR	NCR	%
Idexx - Bovine Rhinotracheitis Virus (BHV-1) gE Antibody Test Kit	8	80	80	100
ID.VET - ID SCREEN® IBR GE COMPETITION	3	30	29	97
<b>TOTAL</b>	<b>11</b>	<b>110</b>	<b>109</b>	<b>99</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

### Conclusion

In total, the laboratories used two different methods. The first method 'Idexx - Bovine Rhinotracheitis Virus (BHV-1) gE Antibody Test Kit' achieved 100% correctness, which means that 80 correct results were submitted. For the second method 'ID.VET - ID SCREEN® IBR GE COMPETITION', a misinterpretation was entered for one sample. The overall score for this method was 97% (29 correct results), which is still higher than the score of 90% that should at least be achieved.

## 2.1.5 AUJESZKY'S DISEASE (AUJ)

### 2.1.5.1 Serology on serum gB

The panel consisted of 7 different samples. Samples PS4, NS1 and NS2 were repeated twice. Therefore, in total, the panel consisted of 10 samples (5 positive and 5 negative samples).

### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS	1 (6)	4 POS 1 NEG 1 NI
PS2	POS	1 (6)	6 POS
PS3	POS	1 (6)	6 NEG
PS4	POS	2 (12)	12 POS
NS1	NEG	2 (12)	12 NEG
NS2	NEG	2 (12)	12 NEG
NS3	NEG	1 (6)	6 NEG

(POS = positive; NEG = negative; NI = not interpreted)

## Used method

Method		Short or long incubation protocol	N	NR	NCR	%
ELISA Competition	Thermofisher Scientific - PrioCHECK® PRV gB	Short	4	40	39	98
ELISA Indirect	Idexx - Pseudorabies Virus gB Antibody Test Kit	Long	2	20	20	100
<b>TOTAL</b>			<b>6</b>	<b>60</b>	<b>58</b>	<b>97</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

## Conclusion

In 2022, six laboratories participated in proficiency test of Aujeszky disease (serum gB) organized by Sciensano. Three methods, PrioCHECK® PRV gB from Thermofisher Scientific, Pseudorabies Virus gB Antibody Test Kit from idexx and PRV/ADV gB Ab from idexx, were selected by the laboratories for the detection of antibodies to the Aujeszky disease virus gB antigen. Two methods fall under the ELISA blocking (competitive) format and one under the indirect format.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. Despite the fact that two laboratories gave incorrect answers for the PS1 sample, all the laboratories achieved a satisfactory performance (> 90%) for the detection of AUJgB-specific antibodies in serum samples.

### 2.1.5.2 Serology on serum gE

The panel consisted of 6 different samples. Samples PS3, PS4, NS1 and NS2 were repeated twice. Therefore, in total, the panel consisted of 10 samples (6 positive and 4 negative samples).

One lab had chosen to test two different methods on the same samples, implying that there were two datasets submitted. These additional results are included in the tables below.

## Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS	1 (10)	10 POS
PS2	POS	1 (10)	10 POS
PS3	POS	2 (20)	20 POS
PS4	POS	2 (20)	20 POS
NS1	NEG	2 (20)	20 NEG
NS2	NEG	2 (20)	20 NEG

(POS = positive; NEG = negative)

## Used method

Method		Short or long incubation protocol	N	NR	NCR	%
ELISA Competition	Idexx - PRV/ADV gl Ab (= PRV/ADV gE)	Short	6	60	60	100
ELISA Competition	Idexx - PRV/ADV gl Ab (= PRV/ADV gE)	Long	1	10	10	100
ELISA Competition	Idexx - PRV/ADV gl Ab (= PRV/ADV gE)	<i>Not applicable</i>	2	20	20	100
ELISA Competition	Thermofisher Scientific - PrioCHECK PRV gE 2.0	Long	1	10	10	100
<b>TOTAL</b>			<b>10</b>	<b>100</b>	<b>100</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

## Conclusion

In 2022, nine laboratories participated in proficiency test of Aujeszky disease (serum gE) organized by Sciensano. Two methods, PRV/ADV gl Ab from Idexx and PrioCHECK PRV gE 2.0 from Thermofisher Scientific, were selected by the laboratories for the detection of antibodies to the Aujeszky disease virus gl antigen (gE). Both methods fall under the ELISA blocking (competitive) format. A distinction was made in the 'PRV/ADV gl Ab' method as the incubation protocol was different or not applicable. One laboratory entered 2 datasets making a total of 10 datasets. In conclusion, both methods achieved a 100% correctness, which implies that 100 correct results were submitted.

## 2.1.6 ENZOOTIC BOVINE LEUKOSIS (EBL)

### 2.1.6.1 Serology on serum

The panel consisted of 7 different samples. Samples PS3, NS1 and NS2 were repeated twice. Therefore, in total, the panel consisted of 10 samples (6 positive and 4 negative samples).

## Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS	1 (6)	6 POS
PS2	POS	1 (6)	6 POS
PS3	POS	2 (12)	12 NEG
PS4	POS	1 (6)	6 POS
PS5	POS	1 (6)	6 NEG
NS1	NEG	2 (12)	12 NEG
NS2	NEG	2 (12)	12 NEG

(POS = positive; NEG = negative)

## Used method

Method		Short or long incubation protocol	N	NR	NCR	%
ELISA Competition	Idexx - Leukosis Blocking Ab test	Short	2	20	20	100
ELISA Competition	ID.VET - ID Screen® BLV Competition	Short	2	20	20	100
ELISA Indirect	Idexx – Indirect ELISA test	Short	2	20	20	100
<b>TOTAL</b>			<b>6</b>	<b>60</b>	<b>60</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

## Conclusion

In 2022, six laboratories participated in proficiency test of enzootic bovine leukosis (EBL) (serology serum) organized by Sciensano. Three methods, Leukosis Blocking Ab test from Idexx, ID Screen® BLV Competition from ID.VET and Indirect ELISA test from Idexx, were selected by the laboratories for the detection of EBL-specific antibodies in serum of ruminants. Two methods fall under the ELISA blocking (competitive) format and one under the indirect format.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. Nevertheless, all the laboratories achieved a satisfactory performance (> 90%).

### 2.1.7 BOVINE VIRAL DIARRHEA (BVD)

#### 2.1.7.1 Serology on serum (ELISA Ab)

The panel consisted of 9 different samples. Negative sample N3 was repeated twice. Therefore, in total, the panel consisted of 10 samples (5 positive and 5 negative samples).

One lab had chosen to test two different methods on the same samples, implying that there were two datasets submitted. These additional results are included in the tables below.



Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
P1	POS	1 (8)	8 POS
P2	POS	1 (8)	7 POS 1 NI
P3	POS	1 (8)	8 POS
P4	POS	1 (8)	8 POS
P5	POS	1 (8)	8 POS
N1	NEG	1 (8)	8 NEG
N2	NEG	1 (8)	8 NEG
N3	NEG	2 (16)	16 NEG
N4	NEG	1 (8)	8 NEG

(POS = positive; NEG = negative, NI = not interpretable)

Used method

Method		Short or long incubation protocol	N	NR	NCR	%
ELISA Indirect	Bio-X Diagnostics - Monoscreen Ab ELISA BVD	Short	2	20	20	100
ELISA Indirect	Idexx - BVD Total Ab	Short	1	10	9	90
ELISA Competition	ID.VET - Idscreen BVD p80 antibody competition	Short	3	30	30	100
ELISA Competition	Bio-X Diagnostics - Monoscreen Ab ELISA BVD	Short	1	10	10	100
ELISA Competition	Thermofisher - BVDV Ab ref 7588940	Short	1	10	10	100
<b>TOTAL</b>			<b>8</b>	<b>80</b>	<b>79</b>	<b>99</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results.)

## Conclusion

In 2022, seven laboratories participated in proficiency test of BVD serology (serum - ELISA) organized by Sciensano. Two indirect ELISA and three blocking ELISA methods were selected by the laboratories for the detection of antibodies against BVD in serum.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. Only the method of Idexx - BVD Total Ab failed to achieve a total score of 100%. The laboratory mentioned on the misreported sample that the sample was quite near to cut off to not interpretable. Also they mentioned that in the daily routine they would have sent it to the Reference Laboratory. Nevertheless, this laboratory obtained a score of 90% which is still in agreement with the guidelines. To conclude; an overall score of 99% was achieved implying that all the five methods used are suitable options for antibody detection against BVD in serum.

### 2.1.7.2 Virology on serum (ELISA Ab)

The panel consisted of 9 different samples. Negative sample N2 was repeated twice. Therefore, in total, the panel consisted of 10 samples (5 positive and 5 negative samples).

#### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
P1	POS	1 (6)	6 POS
P2	POS	1 (6)	6 POS
P3	POS	1 (6)	6 POS
P4	POS	1 (6)	6 POS
P5	POS	1 (6)	6 POS
N1	NEG	1 (6)	6 NEG
N2	NEG	2 (12)	12 NEG
N3	NEG	1 (6)	6 NEG
N4	NEG	1 (6)	6 NEG

(POS = positive; NEG = negative)

### Used method

Method		Short or long incubation protocol	Formula	N	NR	NCR	%
ELISA Indirect	Idexx - Bovine Viral Diarrhoea Virus (BVDV) Antigen Test Kit/Serum Plus	Short	$(OD_{\text{sample}} - OD_{\text{NC}}) / (OD_{\text{PC}} - OD_{\text{NC}})$	2	20	20	100
ELISA Indirect	Idexx - Bovine Viral Diarrhoea Virus (BVDV) Antigen Test Kit/Serum Plus	Short	Sample OD - Negative control mean OD	4	40	40	100
<b>TOTAL</b>				<b>6</b>	<b>60</b>	<b>60</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

### Conclusion

In 2022, six laboratories participated in proficiency test of BVD virology (serum - ELISA) organized by Sciensano. The method Bovine Viral Diarrhoea Virus (BVDV) Antigen Test Kit/Serum Plus from Idexx was selected by the participants. Only there was a difference in the procedure (another formula was used), therefore a distinction between these two was made.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test. As a results, it can be concluded that the method from Idexx is a suitable option for antibody detection against BVD in serum.

#### 2.1.7.3 Virology on serum (RT-qPCR)

The panel consisted of 8 different samples. Negative samples N1 and N2 were repeated twice. Therefore, in total, the panel consisted of 10 samples (5 positive and 5 negative samples).

One lab had chosen to test two different methods on the same samples, implying that there were two datasets submitted. These additional results are included in the tables below.

### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
P1	POS	1 (8)	7 POS 1 NEG
P2	POS	1 (8)	8 POS 1 NEG
P3	POS	1 (8)	6 POS 2 NEG
P4	POS	1 (8)	6 POS 2 NEG
P5	POS	1 (8)	7 POS 1 NEG
N1	NEG	2 (16)	15 NEG 1 POS

Sample ID	Status	Number of repetitions (total results)	Observed result
N2	NEG	2 (16)	15 NEG 1 POS
N3	NEG	1 (8)	8 NEG

(POS = positive; NEG = negative)

#### Used method

Manufacturer extraction protocol / kit	Name extraction protocol / kit	RT-qPCR protocol / kit	N	NR	NCR	%
Qiagen	QIAamp DNA Mini kit	Home made	2	20	15	75
Indical	IndiMag Pathogen Kit	Kit Thermofisher BVD4ALL	2	20	16	80
ThermoFisher Scientific	MagMAX CORE nucleic acid purification kit	Thermofisher vetMAX BVDV screening kit	2	20	20	100
BioX-Adiagene	ADIAMAG XL	Adiavet BVD real time (protocole court) 10K4TRI94	1	10	10	100
<b>TOTAL</b>			<b>7</b>	<b>70</b>	<b>61</b>	<b>87</b>

#### Conclusion

In 2022, seven laboratories participated in proficiency test of BVD virology (serum – RT-qPCR) organized by Sciensano. Different methods were selected by the participants for the identification of the BVD virus in serum of cattle.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. Two laboratories did not achieve the minimum score of 90%. For one laboratory, this was partly due to two coding errors in the Toolkit and partly because they used two methods, one for the detection of BVD I and another for the detection of BVD II. For the second laboratory, an explanation could be found for the poor score of 60%. This laboratory inadvertently entered BVD blood RT-qPCR results instead of their BVD serum RT-qPCR results. After the lab was informed of this, they were able to prove they did have the correct answers, which implies that we can conclude that this is not a bad way of working or that the method is not suitable for this test. Since this concerns a coding error and not an analysis error, this lab does not have to take any further action. However, according to the quality guidelines, they do have to report this in their quality system.

Unlike the two laboratories discussed above, all other laboratories achieved the maximum score of 100%.

#### 2.1.7.4 Virology on blood (ELISA Ab)

The panel consisted of 8 different samples. Positive sample P2 and negative sample N3 were repeated twice. Therefore, in total, the panel consisted of 10 samples (5 positive and 5 negative samples).

##### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
P1	POS	1 (5)	5 POS
P2	POS	2 (10)	10 POS
P3	POS	1 (5)	5 POS
P4	POS	1 (5)	5 POS
N1	NEG	1 (5)	5 NEG
N2	NEG	1 (5)	5 NEG
N3	NEG	2 (10)	10 NEG
N4	NEG	1 (5)	5 NEG

(POS = positive; NEG = negative)

##### Used method

Method		Short or long incubation protocol	Formula	N	NR	NCR	%
ELISA Indirect	Idexx - Bovine Viral Diarrhoea Virus (BVDV) Antigen Test Kit/Serum Plus	Short	$(OD_{\text{sample}} - OD_{\text{NC}}) / (OD_{\text{PC}} - OD_{\text{NC}})$	1	10	10	100
ELISA Indirect	Idexx - Bovine Viral Diarrhoea Virus (BVDV) Antigen Test Kit/Serum Plus	Short	Sample OD - Negative control mean OD	4	40	40	100
<b>TOTAL</b>				<b>5</b>	<b>50</b>	<b>50</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

##### Conclusion

In 2022, five laboratories participated in proficiency test of BVD virology (blood - ELISA) organized by Sciensano. The method Bovine Viral Diarrhoea Virus (BVDV) Antigen Test Kit/Serum Plus from Idexx was selected by the participants. Only there was a difference in the procedure (another formula was used), therefore a distinction between these two was made.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test. As a results, it can be concluded that the method from Idexx is a suitable option for antibody detection against BVD in blood of cattle.

### 2.1.7.5 Virology on blood (RT-qPCR)

The panel consisted of 8 different samples. Positive sample P1 and negative sample N2 were repeated twice. Therefore, in total, the panel consisted of 10 samples (5 positive and 5 negative samples).

#### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
P1	POS	2 (12)	12 POS
P2	POS	1 (6)	6 POS
P3	POS	1 (6)	6 POS
P4	POS	1 (6)	6 POS
N1	NEG	1 (6)	6 NEG
N2	NEG	2 (12)	12 NEG
N3	NEG	1 (6)	6 NEG
N4	NEG	1 (6)	6 NEG

(POS = positive; NEG = negative)

#### Used method

Manufacturer extraction protocol / kit	Name extraction protocol / kit	RT-qPCR protocol / kit	N	NR	NCR	%
Qiagen	RNEASY mini kit	Home made	1	10	10	100
Qiagen	QIAamp DNA Mini kit	Home made	1	10	10	100
Indical	IndiMag Pathogen Kit	Kit Thermofisher BVD4all	2	20	20	100
ThermoFisher Scientific	ThermoFisher Scientific - MagMaxCore	ThermoFisher Scientific - VetMAX BVDV4ALL	2	20	20	100
<b>TOTAL</b>			<b>6</b>	<b>60</b>	<b>60</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

## Conclusion

In 2022, six laboratories participated in proficiency test of BVD virology (blood – RT-qPCR) organized by Sciensano. Four different methods, from Qiagen, Indical and ThermoFisher Scientific were selected by the participants for the identification of the BVD virus in blood of cattle.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test. As a results, it can be concluded that these methods are suitable options the identification of the BVD virus in blood of cattle.

### 2.1.7.6 Virology on ear notch (ELISA Ab)

The panel consisted of 10 different samples. No repetitions were included. Therefore, in total, the panel consisted of 10 samples (5 positive and 5 negative samples).

#### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
P1	POS	1 (6)	6 POS
P2	POS	1 (6)	6 POS
P3	POS	1 (6)	6 POS
P4	POS	1 (6)	6 POS
P5	POS	1 (6)	6 POS
N1	NEG	1 (6)	6 NEG
N2	NEG	1 (6)	6 NEG
N3	NEG	1 (6)	6 NEG
N4	NEG	1 (6)	6 NEG
N5	NEG	1 (6)	6 NEG

(POS = positive; NEG = negative)

#### Used method

Method		Short or long incubation protocol	N	NR	NCR	%
ELISA Indirect	Idexx - Bovine Viral Diarrhoea Virus (BVDV) Antigen Test Kit/Serum Plus	Long	2	20	20	100
ELISA Indirect	Idexx - Bovine Viral Diarrhoea Virus (BVDV) Antigen Test Kit/Serum Plus	Short	4	40	40	100
<b>TOTAL</b>			<b>6</b>	<b>60</b>	<b>60</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

## Conclusion

In 2022, six laboratories participated in proficiency test of BVD virology (ear notch - ELISA) organized by Sciensano. The method Bovine Viral Diarrhoea Virus (BVDV) Antigen Test Kit/Serum Plus from Idexx was selected by all the participants for the detection of antibodies against BVD in ear notch.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test. As a results, it can be concluded that the method from Idexx is a suitable option for antibody detection against BVD in ear notch.

### 2.1.7.7 Virology on ear notch (RT-qPCR)

The panel consisted of 10 different samples. No repetitions were included. Therefore, in total, the panel consisted of 10 samples (5 positive and 5 negative samples).

#### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
P1	POS	1 (6)	6 POS
P2	POS	1 (6)	6 POS
P3	POS	1 (6)	6 POS
P4	POS	1 (6)	6 POS
P5	POS	1 (6)	6 POS
N1	NEG	1 (6)	5 NEG 1 POS
N2	NEG	1 (6)	6 NEG
N3	NEG	1 (6)	6 NEG
N4	NEG	1 (6)	6 NEG
N5	NEG	1 (6)	6 NEG

(POS = positive; NEG = negative)



### Used method

Manufacturer extraction protocol / kit	Name extraction protocol / kit	RT-qPCR protocol / kit	N	NR	NCR	%
Qiagen	RNEASY mini kit	Home made	1	10	10	100
Indical	IndiMag Pathogen Kit	Thermofisher - LSIVETMAX BVD4ALL	1	10	10	100
IDVET	Direct lysis buffer	Virotype BVDV RT-PCR kit	1	10	10	100
BioX-Adiagene	ADIAMAG XL	Adiavet BVD RealTime	1	10	10	100
ThermoFisher Scientific	MagMAX™ CORE Nucleic Acid Purification Kit	LSIVETMAX BVD4ALL	2	20	19	95
Idexx	RealPCR Rapid Lysis Buffer	Home made	1	10	10	100
<b>TOTAL</b>			<b>7</b>	<b>70</b>	<b>69</b>	<b>99</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

### Conclusion

In 2022, seven laboratories participated in proficiency test of BVD virology (ear notch – RT-qPCR) organized by Sciensano. Different methods were selected by the participants for the detection of antibodies against BVD in ear notch.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the minimum score (90%) for this test.

### 2.1.8 VISNA MAEDI (VM)

#### 2.1.8.1 Serology on serum

The panel consisted of 10 different samples. No repetitions were included. Therefore, in total, the panel consisted of 10 samples (4 positive and 6 negative samples).

### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS	1 (4)	4 POS
PS2	POS	1 (4)	4 POS
PS3	POS	1 (4)	4 POS
PS4	POS	1 (4)	4 POS
NS1	NEG	1 (4)	4 NEG
NS2	NEG	1 (4)	4 NEG
NS3	NEG	1 (4)	4 NEG
NS4	NEG	1 (4)	4 NEG
NS5	NEG	1 (4)	4 NEG
NS6	NEG	1 (4)	4 NEG

(POS = positive; NEG = negative)

### Used method

Method		Short or long incubation protocol	N	NR	NCR	%
ELISA Indirect	ID.VET - ID Screen MVV/CAEV Indirect	Not applicable	2	20	20	100
ELISA Indirect	Hyphen Biomed - ELITEST MVV/CAEV	Short	2	20	20	100
<b>TOTAL</b>			<b>4</b>	<b>40</b>	<b>40</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

### Conclusion

In 2022, four laboratories participated in proficiency test of Visna Maedi serology (serum) organized by Sciensano. The method ID Screen MVV/CAEV Indirect from ID.VET and ELITEST MVV/CAEV from Hyphen Biomed were selected by the participants for the detection of antibodies against the Visna Maedi virus in serum. These methods fall under the indirect format.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test. As a results, it can be concluded that the methods from ID.VET and Hyphen Biomed are suitable options for antibody detection against the Visna Maedi virus in serum of sheep.

## 2.2 Bacteriology

The samples for the surveys of this section were produced by the Bacteriology laboratory of the Directorate Infectious Diseases in Animals of Sciensano.

### 2.2.1 Q-FEVER (QF)

#### 2.2.1.1 Serology on serum

The panel consisted of 20 different samples, 15 positive and 5 negative samples.

##### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS	4 (20)	20 POS
PS2	POS	3 (15)	15 POS
PS3	POS	4 (20)	20 POS
PS4	POS	2 (10)	10 POS
PS5	POS	2 (10)	10 POS
NS1	NEG	5 (25)	25 POS

(POS = positive; NEG = negative)

##### Used method

Method	N	NR	NCR	% Agreement
ThermoFisher - PrioCheck Ruminant Q Fever Ab Plate Kit	4	80	80	100
ID.VET - ID SCREEN® Q FEVER INDIRECT MULTI-SPECIES	1	20	20	100
<b>TOTAL</b>	<b>5</b>	<b>100</b>	<b>100</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

##### Conclusion

In total, two different methods were used by the laboratories. All these methods achieved 100% correctness, which means that 100 correct results were submitted.

#### 2.2.1.2 Serology on milk

The panel consisted of 20 different samples, 16 positive and 4 negative samples.

##### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS	4 (12)	12 POS
PS2	POS	4 (12)	12 POS
PS3	POS	4 (12)	12 POS
PS4	POS	4 (12)	12 POS
NS1	NEG	4 (12)	12 POS

(POS = positive; NEG = negative)

### Used method

Method	N	NR	NCR	% Agreement
Thermofisher - PrioCheck Ruminant Q Fever Ab Plate Kit	3	60	60	100
<b>TOTAL</b>	<b>3</b>	<b>60</b>	<b>60</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

### Conclusion

Only one method was used by the laboratories. This method achieved 100% correctness, which means that 60 correct results were submitted.

## 2.2.2 BRUCELLOSIS (BRU)

### 2.2.2.1 Serology on milk

The panel consisted of 6 different samples. Samples PM2 and NM2 were repeated twice. Sample PM4 was repeated three times. Samples PM1 and NM1 were repeated four times. Sample PM3 was repeated five times. Therefore, in total, the panel consisted of 20 samples (14 positive and 6 negative samples).

### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PM1	POS	4 (20)	20 POS
PM2	POS	2 (10)	10 POS
PM3	POS	5 (25)	25 POS
PM4	POS	3 (15)	15 POS
NM1	NEG	4 (20)	20 NEG
NM2	NEG	2 (10)	10 NEG

(POS = positive; NEG = negative)

### Used method

Method		Short or long incubation protocol	N	NR	NCR	%
ELISA Indirect	IDEXX - Brucellosis Antibody test kit (Tank milk)	Short	5	25	25	100
<b>TOTAL</b>			<b>5</b>	<b>25</b>	<b>25</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

## Conclusion

In 2022, five laboratories participated in proficiency test of *Brucella* serology (milk) organized by Sciensano. The method Brucellosis Antibody test kit (Tank milk) from IDEXX was selected by all the participants for the detection of antibodies against *Brucella* in milk. This method falls under the indirect format.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test. As a results, it can be concluded that the method from IDEXX is a suitable option for antibody detection against *Brucella* in tank milk.

### 2.2.2.2 Bacteriology on organs

The panel consisted of 10 different samples (5 positive and 5 negative samples). No repetitions of samples were included in this panel.

#### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PO1	POS	1 (4)	4 POS
PO2	POS	1 (4)	3 POS 1 NEG
PO3	POS	1 (4)	4 POS
PO4	POS	1 (4)	4 POS 1 NEG
PO5	POS	1 (4)	4 POS
NO1	NEG	1 (4)	4 NEG
NO2	NEG	1 (4)	4 NEG
NO3	NEG	1 (4)	4 NEG
NO4	NEG	1 (4)	4 NEG
NO5	NEG	1 (4)	4 NEG

(POS = positive; NEG = negative)

#### Used method

Reagens	Batchnummer	N	NR	NCR	%
1) Farell - home made	1) PHVAN/22/04	1	10	10	100
2) Oxydase - Sigma	2) MKCC4915				
3) Reagent Urease - home made	3) PAMIC/22/03				
4) Serum for agglutination anti-S - REMEL	4) PAMIC/22/01				
5) Serum for agglutination anti-R - ANSES	5) PAMIC/22/01				
6) Negative serum for agglutination - ANSES	6) PAMIC/22/01				
7) Merck - H <sub>2</sub> O <sub>2</sub> 30%	7) K54376510222				
1) Remel - Agglutination serum B. abortus	1) 3324198	1	8	10	80
2) S�rum Agglutination B. melitensis Remel	2) 3338873				
3) Oxydase Bactident Sigma Aldrich	3) HC297883				

Reagents	Batchnummer	N	NR	NCR	%
Homemade medium - BRU22/22	/	1	10	10	100
Anses - homemade	/	1	10	10	100
<b>TOTAL</b>		<b>4</b>	<b>38</b>	<b>40</b>	<b>95</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

### Conclusion

In 2022, four laboratories participated in the proficiency test for *Brucella* bacteriology (organs) organized by Sciensano. Different reagents were selected by the participants for the isolation and identification of *Brucella* in organs.

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 the reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. Three laboratories succeeded in achieving the maximum score (100%) for this test. Only one laboratory had a score of 80% and did not achieve the 90% standard. This can be explained because the methodology performed in this laboratory does not allow to identify *B. ovis* and/or *B. canis* and the two failed samples were spiked with *B. canis*. Therefore, according to their procedures, it is consistent that they cannot detect or identify *B. canis*.

## 2.2.3 BOVINE TUBERCULOSIS (BT)

### 2.2.3.1 Serology on serum

The panel consisted of 6 different samples. All samples were repeated at least twice (see table below). Therefore, in total, the panel consisted of 20 samples (15 positive and 5 negative samples).

#### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS/NEG	5 (25)	24 POS 1 NEG
PS2	POS	2 (10)	10 POS
PS3	POS	3 (15)	15 POS
PS4	POS	5 (25)	25 POS
NS1	NEG	3 (15)	15 NEG
NS2	NEG	2 (10)	10 NEG

(POS = positive; NEG = negative)

#### Used method

Method		Short or long incubation protocol	N	NR	NCR	%
ELISA Indirect	IDEXX - Mycobacterium Bovis Antibody Test Kit	Short	5	100	100	100
<b>TOTAL</b>			<b>5</b>	<b>100</b>	<b>100</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

#### Conclusion

In 2022, five laboratories participated in proficiency test of Bovine Tuberculosis serology (serum) organized by Sciensano. The method Mycobacterium Bovis Antibody Test Kit from IDEXX was selected by all the participants for the detection of antibodies against Bovine Tuberculosis in serum of cattle. This method is an indirect ELISA.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test.

#### 2.2.3.2 Gamma interferon on serum/plasma

The panel consisted of 5 different samples. All samples were repeated at least twice (see table below). Therefore, in total, the panel consisted of 20 samples (15 positive and 5 negative samples).

#### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
PG1	POS	4 (16)	16 POS
PG2	POS	5 (20)	20 POS
PG3	POS	6 (24)	24 POS
NG1	NEG	2 (8)	8 NEG
NG2	NEG	3 (12)	12 NEG

(POS = positive; NEG = negative)

#### Used method

Method		Short or long incubation protocol	N	NR	NCR	%
ELISA Indirect	ID.VET - ID Screen ruminant IFN-g	Short	4	80	80	100
<b>TOTAL</b>			<b>4</b>	<b>80</b>	<b>80</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

## Conclusion

In 2022, four laboratories participated in proficiency test of Bovine Tuberculosis gamma interferon (serum) organized by Sciensano. The method ID Screen ruminant IFN-g from ID.VET was selected by all the participants for the detection of gamma Interferon. This method is a sandwich ELISA designed to catch the gamma interferon produced in the tested plasmas.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test.

## 2.2.4 LEPTOSPIROSIS (LEPT)

### 2.2.4.1 Bacteriology on organs

The panel consisted of 3 different samples. Positive samples OP1 and OP2 were repeated twice. Therefore, in total, the panel consisted of 5 samples (4 positive and 1 negative sample).

Unfortunately, one laboratory did not submit its results even after sending a reminder to them. Therefore, the table below shows the results of three laboratories instead of four.

#### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
OP1	POS	2 (6)	6 POS
OP2	POS	2 (6)	6 POS
ON1	NEG	1 (3)	3 NEG

(POS = positive; NEG = negative)

#### Used method

Manufacturer extraction protocol / kit	Name extraction protocol / kit	RT-qPCR protocol / kit	N	NR	NCR	%
ThermoFisher Scientific	Kit MagMax	Homemade (SOP/BAC/ANA16)	1	10	10	100
Indical	IndiMag Pathogen Kit	Thermofisher Vetmax SARP kit	1	10	10	100
Indical	IndiMag Pathogen Kit	Ingenetix - BactoReal Kit Leptospirosis	1	10	10	100
<b>TOTAL</b>			<b>3</b>	<b>30</b>	<b>30</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)



## Conclusion

In 2022, three laboratories participated in proficiency test of Leptospirosis bacteriology (organs) organized by Sciensano. Unfortunately, the fourth laboratory that was registered for this PT did not submit their results, even not after a reminder of the deadline via mail.

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 reference laboratory of the Scientific Directorate Infectious Diseases in Animals of Sciensano. Different methods, from ThermoFisher Scientific and Indical were selected by the participants for the detection of pathogenic *Leptospira* spp. bacteria in organs. All laboratories succeeded in achieving the maximum score (100%) for this test.

## **2.2.5 SALMONELLA PULLORUM-GALLINARUM**

### **2.2.5.1 Bacteriology on organs**

The panel consisted of 5 different samples. On the one hand, positive samples P01, P03 and P04 were repeated twice and P02 was once in the panel. On the other hand, negative sample N01 was repeated three times. Therefore, in total, the panel consisted of 10 samples (7 positive and 3 negative samples).

#### Results per sample

Sample ID	Status	Number of repetitions (total results)	Observed result
P01	POS	2 (6)	6 POS
P02	POS	1 (3)	3 POS
P03	POS	2 (6)	6 POS
P04	POS	2 (6)	6 POS
N01	NEG	3 (9)	9 NEG

(POS = positive; NEG = negative)

## Used method

Method	Reagent(s)	Batch number(s)	N	NR	NCR	%
Method suitable for detecting non-motile <i>Salmonella</i> spp.	1. Bio-Rad - Peptoned water 2. Bio-Rad - RVS 3. Other - other - Oxoid/BGA 4. Bio-Rad - Rapid SALM Agar 5. Thermofisher - Lysine 6. Bio-Trading/TSI 7. Sorbitol/mobilité: home-made 8. Dulcitol: home-made	1. 64478316 2. 64495052 3. 4385004 4. 64508761 5. 3557567 6. 2225005925 7. / 8. /	1	10	10	100
Method suitable for detecting non-motile <i>Salmonella</i> spp.	1. Bio-Rad – RVS 2. Thermofisher milieu BGA 3. Biorad milieu RapidSalm	1. 64474883 2. 4386770 3. 64515253	1	10	10	100
Method suitable for detecting non-motile <i>Salmonella</i> spp.	1. Bio-Rad - RVS 2. Thermofisher - BGA agar 3. Thermofisher - brilliance salmonella agar 4. Biomerieux - BPW	1. 64495052 2. 2283409 3. 2298746 4. 2149350	1	10	10	100
<b>TOTAL</b>			<b>3</b>	<b>30</b>	<b>30</b>	<b>100</b>

(N= number of laboratories; NR = number of results; NCR = number of correct results)

## Conclusion

In 2022, three laboratories participated in proficiency test of *Salmonella* bacteriology (organs) organized by Sciansano. According to the procedure currently in force, the performance of a participating laboratory is satisfactory when no mistakes are detected (100% of agreement) for strong positive samples. In the case of weak positive and negative samples, one error is allowed (90% of agreement). All laboratories succeeded in achieving the maximum score (100%) for this test.

### 3 GENERAL EVALUATION

#### 3.1 Summary of results

Below you can find a table (Table VI) with all the results obtained by the organized PTs in 2022. A total score of 99% was achieved.

**Table VI.** Summary of the results (NP= number of participants; NR= number of results; NCR= number of correct results).

Name of proficiency test			Concerned methods	NP	NR	NCR	%
<b>Porcine Reproductive and Respiratory Syndrome</b>	Serology	Serum	ELISA (Ab)	7	126	126	100
	Serology	Serum	ELISA (Ab)	5	100	100	100
<b>Q-Fever</b>	Serology	Milk	ELISA (Ab)	3	60	60	100
	Serology	Serum	ELISA (Ab)	6	60	60	100
<b>African Swine Fever (type II strain)</b>	Virology	Serum	ELISA (Ab)	6	60	60	100
	Serology	Serum	ELISA (Ab)	5	100	100	100
<b>Classical Swine Fever</b>	Serology	Serum	ELISA (Ab)	5	100	100	100
<b>Infectious Bovine Rhinotracheitis</b>	Serology	Serum gB	ELISA (Ab)	9	90	90	100
	Serology	Serum gE	ELISA (Ab)	11	110	109	99
<b>Aujeszky's Disease</b>	Serology	Serum gB	ELISA (Ab)	6	60	58	98
	Serology	Serum gE	ELISA (Ab)	10	100	100	100
<b>Enzootic Bovine Leukosis</b>	Serology	Serum	ELISA (Ab)	6	60	60	100
<b>Brucellosis</b>	Serology	Milk	ELISA (Ab)	5	25	25	100
	Bacteriology	Organs	Isolation	4	40	38	95
<b>Bovine Viral Diarrhea</b>	Serology	Serum	ELISA (Ab)	8	80	79	99
	Virology	Serum	ELISA (Ag)	6	60	60	100
	Virology	EDTA-blood	ELISA (Ag)	5	50	50	100
	Virology	Ear notch	ELISA (Ag)	6	60	60	100
	Virology	Serum	RT-qPCR	7	70	61	87
	Virology	EDTA-blood	RT-qPCR	6	60	60	100
	Virology	Ear notch	RT-qPCR	7	70	69	99
<b>Bovine Tuberculosis</b>	Serology	Serum	ELISA (Ab)	5	100	100	100
	Gamma interferon	Serum	ELISA (Ab)	4	80	80	100
<b>Leptospirosis</b>	Bacteriology	Organs	Isolation	3	30	30	100
<b>Visna Maedi Virus</b>	Serology	Serum	ELISA (Ab)	4	40	40	100
<b>Salmonella Pullorum/Gallinarum</b>	Bacteriology	Organs	Isolation	3	30	30	100
<b>TOTAL</b>					<b>1 721</b>	<b>1 705</b>	<b>99</b>

## 3.2 Analysis of the incorrect results

The encountered problems were summarized in Table VII. The cause of the problem can be diverse and can sometimes be identified.

**Table VII.** Analysis of the incorrect results.

Name of proficiency test			Concerned methods	Explanation
<b>IBR</b>	Serology	Serum gE	ELISA (Ab)	Unknown (one lab entered an incorrect result, but the minimal score of 90% was achieved therefore no actions were taken)
<b>AUJ</b>	Serology	Serum gB	ELISA (Ab)	Unknown (one lab entered an incorrect result, but the minimal score of 90% was achieved therefore no actions were taken)
<b>BRU</b>	Bacteriology	Organs	Isolation	One lab entered two incorrect results (methodology performed by the lab does not allow to identify <i>B. ovis</i> and/or <i>B. canis</i> ).
<b>BVD</b>	Serology	Serum	ELISA (Ab)	The sample was quite near to cut off and therefore this lab entered 'not interpretable' as a result. The minimal score of 90% was achieved therefore no actions were taken.
	Virology	Serum	RT-qPCR	Lab 1: two coding errors in the Toolkit Lab 2: inadvertently switched results (= coding error).
	Virology	Ear notch	RT-qPCR	Unknown (one lab entered an incorrect result, but the minimal score of 90% was achieved therefore no actions were taken)

## 3.3 General conclusions

- Initially, a PT virology was foreseen whereby it was intended to detect the PRRS virus by the use of the RT-qPCR method. Unfortunately this PT could not be conducted this year due to staff shortages caused by the covid crisis. This PT will be organized in 2023.
- A PT BVD serology on milk was foreseen, but this PT will be disappear from the calendar as there are no labs in Belgium (and only a few in Europe) that perform this analysis.
- The laboratories achieved a total score of 99%.
- If laboratories had a no satisfactory score, it was mostly due to coding errors as shown in Table VII.

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END

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