

**BIOLOGICAL HEALTH RISKS  
QUALITY OF LABORATORIES**

**COMMITTEE OF EXPERTS**

**EXTERNAL QUALITY ASSESSMENT  
IN VETERINARY DIAGNOSIS**

**DEFINITIVE GLOBAL REPORT  
VETERINARY MEDICINE  
BOVINE VIRAL DIARRHEA (BVD)  
PROFICIENCY TEST 2023/10**

**Sciensano/PT VET BVD/2023-10/E**

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

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

<b>COMMITTEE OF EXPERTS</b>
-----------------------------

<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>		
Gaëtan De Gryse	<b>Sciensano</b>		

A draft version of this report was submitted to the expert(s) on 17/01/2024.

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

**Date of publication:** 24/01/2024

All the reports are available on our webpage:

- 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>

# TABLE OF CONTENTS

<b>1</b>	<b>INTRODUCTION .....</b>	<b>5</b>
<b>2</b>	<b>AIM .....</b>	<b>5</b>
<b>3</b>	<b>MATERIALS AND METHODS .....</b>	<b>5</b>
<b>3.1</b>	<b>Serology (serum - ELISA) .....</b>	<b>5</b>
3.1.1	The participants .....	5
3.1.2	The samples .....	5
3.1.3	Homogeneity.....	6
3.1.4	Target values .....	6
3.1.5	Stability .....	6
3.1.6	Randomisation and panel composition .....	6
3.1.7	Threshold for qualification.....	7
<b>3.2</b>	<b>Virology (serum - ELISA) .....</b>	<b>7</b>
3.2.1	The participants .....	7
3.2.2	The samples .....	7
3.2.3	Homogeneity.....	8
3.2.4	Target values .....	8
3.2.5	Stability .....	8
3.2.6	Randomisation and panel composition .....	8
3.2.7	Threshold for qualification.....	9
<b>3.3</b>	<b>Virology (serum - RT-qPCR) .....</b>	<b>9</b>
3.3.1	The participants .....	9
3.3.2	The samples .....	9
3.3.3	Homogeneity.....	9
3.3.4	Target values .....	9
3.3.5	Stability .....	10
3.3.6	Randomisation and panel composition .....	10
3.3.7	Threshold for qualification.....	10
<b>3.4</b>	<b>Virology (blood - ELISA) .....</b>	<b>10</b>
3.4.1	The participants .....	10
3.4.2	The samples .....	11
3.4.3	Homogeneity.....	11
3.4.4	Target values .....	11
3.4.5	Stability .....	11
3.4.6	Randomisation and panel composition .....	12
3.4.7	Threshold for qualification.....	12
<b>3.5</b>	<b>Virology (blood - RT-qPCR) .....</b>	<b>12</b>
3.5.1	The participants .....	12
3.5.2	The samples .....	13
3.5.3	Homogeneity.....	13
3.5.4	Target values .....	13
3.5.5	Stability .....	13
3.5.6	Randomisation and panel composition .....	14
3.5.7	Threshold for qualification.....	14
<b>3.6</b>	<b>Virology (ear notch - ELISA) .....</b>	<b>15</b>
3.6.1	The participants .....	15
3.6.2	The samples .....	15
3.6.3	Homogeneity.....	15
3.6.4	Target values .....	15
3.6.5	Stability .....	16
3.6.6	Randomisation and panel composition .....	16
3.6.7	Threshold for qualification.....	16
<b>3.7</b>	<b>Virology (ear notch - RT-qPCR).....</b>	<b>16</b>
3.7.1	The participants .....	16
3.7.2	The samples .....	17
3.7.3	Homogeneity.....	17
3.7.4	Target values .....	17

3.7.5	Stability .....	17
3.7.6	Randomisation and panel composition .....	18
3.7.7	Threshold for qualification .....	18
<b>4</b>	<b>TIMELINE .....</b>	<b>18</b>
<b>5</b>	<b>RESULTS .....</b>	<b>19</b>
<b>5.1</b>	<b>Serology (serum - ELISA) .....</b>	<b>19</b>
5.1.1	Results per sample .....	19
5.1.2	Used ELISA protocol/kit .....	19
5.1.3	Conclusion .....	19
<b>5.2</b>	<b>Virology (serum - ELISA) .....</b>	<b>20</b>
5.2.1	Results per sample .....	20
5.2.2	Used ELISA protocol/kit .....	20
5.2.3	Conclusion .....	20
<b>5.3</b>	<b>Virology (serum – RT-qPCR) .....</b>	<b>21</b>
5.3.1	Results per sample .....	21
5.3.2	Used RT-PCR protocol/kit.....	21
5.3.3	Used extraction protocol .....	22
5.3.4	Conclusion .....	22
<b>5.4</b>	<b>Virology (blood – ELISA).....</b>	<b>23</b>
5.4.1	Results per sample .....	23
5.4.2	Used ELISA protocol/kit .....	23
5.4.3	Conclusion .....	23
<b>5.5</b>	<b>Virology (blood - RT-qPCR) .....</b>	<b>24</b>
5.5.1	Results per sample .....	24
5.5.2	Used RT-PCR protocol/kit.....	24
5.5.3	Used extraction protocol .....	25
5.5.4	Conclusion .....	25
<b>5.6</b>	<b>Virology (ear notch - ELISA).....</b>	<b>26</b>
5.6.1	Results per sample .....	26
5.6.2	Used ELISA protocol/kit .....	26
5.6.3	Conclusion .....	26
<b>5.7</b>	<b>Virology (ear notch - RT-qPCR).....</b>	<b>27</b>
5.7.1	Results per sample .....	27
5.7.2	Used RT-PCR protocol/kit.....	27
5.7.3	Used extraction protocol .....	28
5.7.4	Conclusion .....	28
<b>6</b>	<b>ANNEXES (NOT UNDER ACCREDITATION) .....</b>	<b>29</b>
<b>6.1</b>	<b>Annex 1: Quantitative results .....</b>	<b>29</b>
6.1.1	Serology serum (ELISA) .....	29
6.1.2	Virology serum (ELISA) .....	32
6.1.3	Virology blood (ELISA).....	33
<b>6.2</b>	<b>Annex 2: Additional information .....</b>	<b>34</b>

# 1 INTRODUCTION

Details relevant to the proficiency test (PT) are available in the procedure SOP 2.5/01 'Management of the proficiency tests organized by the scientific directorate infectious diseases in animals'. The PT was organized according to the ISO17043 'Conformity assessment - General requirements for proficiency testing' norm.

## 2 AIM

This proficiency test was divided into two different parts: serology and virology:

- The aim of the **serology part** was to evaluate the ability of the participating laboratories to detect the absence or presence of antibodies against BVD in serum of cattle.
- The aim of the **virology part** was to evaluate the ability of the participating laboratories to identify BVD virus in serum, blood and ear notch of cattle.

## 3 MATERIALS AND METHODS

### 3.1 Serology (serum - ELISA)

#### 3.1.1 THE PARTICIPANTS

Eight laboratories participated in the proficiency test of BVD serology on serum (ELISA). The names of the participating laboratories are:

- Sciensano, department of Veterinary Virology
- ARSIA
- Dierengezondheidszorg Vlaanderen (DGZ)
- LAVETAN
- ANSES Unité Pathologie et Bien-être des ruminants (PBER)-Site de Niort
- Poulpharm
- IDEXX Technologies GmbH (Bern)
- Laboratorio Central De Veterinaria (LCV) (ALGETE) M.A.P.A.

#### 3.1.2 THE SAMPLES

The samples (frozen serum) were prepared by the National Reference Laboratory (NRL), VIRENBEE, Sciensano.

Information about the **origin** of the samples:

- Sample PS1: wpos 16.01
- Sample PS2: 24082012CATZELE1
- Sample PS3: 24082012CATZELE3
- Sample NS1: glorieux n°5=51731955

All samples are field samples stored frozen before aliquotation and transport.

### 3.1.3 HOMOGENEITY

The homogeneity of the samples was tested by the NRL on three aliquots (three serum samples, each 250 µL) of each sample using ELISA before and after the PT. The NRL obtained each time the same qualitative result. Therefore, the samples were considered as homogeneous.

### 3.1.4 TARGET VALUES

The target values were determined by the NRL based on the homogeneity tests. The panel consisted of four different samples. However, positive sample PS1 and negative sample NS1 were replicated three times. Additionally, positive samples PS2 and PS3 were repeated two times. Therefore, the panel included ten samples in total.

Sample content	Repetition	Expected result
PT2023BVDAbSER_PS1	3	POS
PT2023BVDAbSER_PS2	2	POS
PT2023BVDAbSER_PS3	2	POS
PT2023BVDAbSER_NS1	3	NEG

(POS = positive; NEG = negative)

### 3.1.5 STABILITY

The stability was determined by comparison of the pre-proficiency test results with the results obtained by the NRL during and after the proficiency test. The samples were considered as stable.

### 3.1.6 RANDOMISATION AND PANEL COMPOSITION

Since a specific number has been assigned to each laboratory, the randomisation has been performed as follows:

Sample content: PT2023 BVDAb SER_	97505	97507	97508	97509
PS1 (1)	BVDSERSER(E)23-1	BVDSERSER(E)23-1	BVDSERSER(E)23-3	BVDSERSER(E)23-2
PS1 (2)	BVDSERSER(E)23-4	BVDSERSER(E)23-7	BVDSERSER(E)23-7	BVDSERSER(E)23-4
PS1 (3)	BVDSERSER(E)23-10	BVDSERSER(E)23-10	BVDSERSER(E)23-8	BVDSERSER(E)23-10
PS2 (1)	BVDSERSER(E)23-7	BVDSERSER(E)23-3	BVDSERSER(E)23-6	BVDSERSER(E)23-5
PS2 (2)	BVDSERSER(E)23-9	BVDSERSER(E)23-6	BVDSERSER(E)23-10	BVDSERSER(E)23-6
PS3 (1)	BVDSERSER(E)23-3	BVDSERSER(E)23-2	BVDSERSER(E)23-5	BVDSERSER(E)23-8
PS3 (2)	BVDSERSER(E)23-6	BVDSERSER(E)23-5	BVDSERSER(E)23-9	BVDSERSER(E)23-9
NS1 (1)	BVDSERSER(E)23-2	BVDSERSER(E)23-4	BVDSERSER(E)23-1	BVDSERSER(E)23-1
NS1 (2)	BVDSERSER(E)23-5	BVDSERSER(E)23-8	BVDSERSER(E)23-2	BVDSERSER(E)23-3
NS1 (3)	BVDSERSER(E)23-8	BVDSERSER(E)23-9	BVDSERSER(E)23-4	BVDSERSER(E)23-7

Sample content: PT2023 BVDAb SER_	97513	97540	97544	97621
PS1 (1)	BVDSERSER(E)23-4	BVDSERSER(E)23-3	BVDSERSER(E)23-4	BVDSERSER(E)23-3
PS1 (2)	BVDSERSER(E)23-6	BVDSERSER(E)23-7	BVDSERSER(E)23-5	BVDSERSER(E)23-4
PS1 (3)	BVDSERSER(E)23-8	BVDSERSER(E)23-9	BVDSERSER(E)23-10	BVDSERSER(E)23-8
PS2 (1)	BVDSERSER(E)23-7	BVDSERSER(E)23-4	BVDSERSER(E)23-2	BVDSERSER(E)23-1
PS2 (2)	BVDSERSER(E)23-10	BVDSERSER(E)23-5	BVDSERSER(E)23-3	BVDSERSER(E)23-2
PS3 (1)	BVDSERSER(E)23-1	BVDSERSER(E)23-8	BVDSERSER(E)23-7	BVDSERSER(E)23-5
PS3 (2)	BVDSERSER(E)23-5	BVDSERSER(E)23-10	BVDSERSER(E)23-9	BVDSERSER(E)23-10
NS1 (1)	BVDSERSER(E)23-2	BVDSERSER(E)23-1	BVDSERSER(E)23-1	BVDSERSER(E)23-6
NS1 (2)	BVDSERSER(E)23-3	BVDSERSER(E)23-2	BVDSERSER(E)23-6	BVDSERSER(E)23-7
NS1 (3)	BVDSERSER(E)23-9	BVDSERSER(E)23-6	BVDSERSER(E)23-8	BVDSERSER(E)23-9

### 3.1.7 THRESHOLD FOR QUALIFICATION

Following the procedure, a participating laboratory is only qualified if the level of agreement for the ten reference samples is at least 90%.

## 3.2 Virology (serum - ELISA)

### 3.2.1 THE PARTICIPANTS

Five laboratories participated in the proficiency test of BVD virology on serum (ELISA). The names of the participating laboratories are:

- Sciensano, department of Veterinary Virology
- ARSIA
- Dierengezondheidszorg Vlaanderen (DGZ)
- LAVETAN
- ANSES Unité Pathologie et Bien-être des ruminants (PBER)-Site de Niort

### 3.2.2 THE SAMPLES

The samples (frozen serum) were prepared by the National Reference Laboratory (NRL), Service of VIRENBEE, Sciensano.

Information about the **origin** of the samples:

- Sample SP1: 16052014 CATSTOFFELS
- Sample SP2: PONCIN SOLANGE
- Sample SN1: 24.08.2012 CATZELE18
- Sample SN2: 24.08.2012 CATZELE6

All samples are field samples stored frozen before aliquotation and transport.

### 3.2.3 HOMOGENEITY

The homogeneity of the samples was tested by the NRL on three aliquots (three serum samples, each 250 µL) of each sample using ELISA before and after the PT. The NRL obtained each time the same qualitative result. Therefore, the samples were considered as homogeneous.

### 3.2.4 TARGET VALUES

The target values were determined by the NRL based on the homogeneity tests. The panel consisted of four different samples. However, positive sample SP1 was replicated two times. Additionally, negative samples SN1 and SN2 were repeated two times. Therefore, the panel included seven samples in total.

Sample content	Repetition	Expected result
PT2023BVDAgVIR_SP1	2	POS
PT2023BVDAgVIR_SP2	1	POS
PT2023BVDAgVIR_SN1	2	POS
PT2023BVDAgVIR_SN2	2	NEG

(POS = positive; NEG = negative)

### 3.2.5 STABILITY

The stability was determined by comparison of the pre-proficiency test results with the results obtained by the NRL during and after the proficiency test. The samples were considered as stable.

### 3.2.6 RANDOMISATION AND PANEL COMPOSITION

Since a specific number has been assigned to each laboratory, the randomisation has been performed as follows:

Sample content: PT2023BVDAgVIR_	97505	97507	97508	97509	97513
SP1 (1)	BVDVIR SER(E)23-1	BVDVIR SER(E)23-6	BVDVIR SER(E)23-2	BVDVIR SER(E)23-2	BVDVIR SER(E)23-3
SP1 (2)	BVDVIR SER(E)23-5	BVDVIR SER(E)23-7	BVDVIR SER(E)23-4	BVDVIR SER(E)23-1	BVDVIR SER(E)23-6
SP2	BVDVIR SER(E)23-2	BVDVIR SER(E)23-5	BVDVIR SER(E)23-7	BVDVIR SER(E)23-3	BVDVIR SER(E)23-5
SN1 (1)	BVDVIR SER(E)23-4	BVDVIR SER(E)23-1	BVDVIR SER(E)23-3	BVDVIR SER(E)23-4	BVDVIR SER(E)23-2
SN1 (2)	BVDVIR SER(E)23-6	BVDVIR SER(E)23-2	BVDVIR SER(E)23-6	BVDVIR SER(E)23-6	BVDVIR SER(E)23-4
SN2 (1)	BVDVIR SER(E)23-3	BVDVIR SER(E)23-3	BVDVIR SER(E)23-1	BVDVIR SER(E)23-5	BVDVIR SER(E)23-1
SN2 (2)	BVDVIR SER(E)23-7	BVDVIR SER(E)23-4	BVDVIR SER(E)23-5	BVDVIR SER(E)23-7	BVDVIR SER(E)23-7



### 3.2.7 THRESHOLD FOR QUALIFICATION

Following the procedure, a participating laboratory is only qualified if the level of agreement for the seven reference samples is 100%.

## 3.3 Virology (serum - RT-qPCR)

### 3.3.1 THE PARTICIPANTS

Seven laboratories participated in the proficiency test of BVD virology on serum (RT-qPCR). The names of the participating laboratories are:

- Sciensano, department of Veterinary Virology
- ARSIA
- Dierengezondheidszorg Vlaanderen (DGZ)
- ANSES Unité Pathologie et Bien-être des ruminants (PBER)-Site de Niort
- Biosellal
- LSI-ThermoFisher Scientific (France)
- Poulpharm

### 3.3.2 THE SAMPLES

The samples (frozen serum) were prepared by the National Reference Laboratory (NRL), Service of VIRENBEE, Sciensano.

Information about the **origin** of the samples:

- Sample SP1: 04.12.12 CATBRAEM 0032
- Sample SP2: 01102012CATRendac1 - DIL 1/1000
- Sample SP3: 16.05.24CATSTOFFELS
- Sample SP4: PONCIN SOLANGE
- Sample SN1: 14082012 CATZELE9

All samples are field samples stored frozen before aliquotation and transport.

### 3.3.3 HOMOGENEITY

The homogeneity of the samples was tested by the NRL on three aliquots (three serum samples, each 500 µL) of each sample using RT-PCR before and after the PT. The NRL obtained each time the same qualitative result. Therefore, the samples were considered as homogeneous.

### 3.3.4 TARGET VALUES

The target values were determined by the NRL based on the homogeneity tests. The panel consisted of five different samples. No repetitions were included.

Sample content	Repetition	Expected result
PT2023BVDVIR_SP1	1	POS
PT2023BVDVIR_SP2	1	POS
PT2023BVDVIR_SP3	1	POS
PT2023BVDVIR_SP4	1	POS
PT2023BVDVIR_SN1	1	NEG

(POS = positive; NEG = negative)

### 3.3.5 STABILITY

The stability was determined by comparison of the pre-proficiency test results with the results obtained by the NRL during and after the proficiency test. The samples were considered as stable.

### 3.3.6 RANDOMISATION AND PANEL COMPOSITION

Since a specific number has been assigned to each laboratory, the randomisation has been performed as follows:

### 3.3.7 THRESHOLD FOR QUALIFICATION

Following the procedure, a participating laboratory is only qualified if the level of agreement for the five reference samples is 100%.

Sample content: PT2023 BVDVIR_	97505	97507	97508	97513
SP1	BVDVIRSER(P)23-5	BVDVIRSER(P)23-2	BVDVIRSER(P)23-2	BVDVIRSER(P)23-2
SP2	BVDVIRSER(P)23-3	BVDVIRSER(P)23-4	BVDVIRSER(P)23-1	BVDVIRSER(P)23-4
SP3	BVDVIRSER(P)23-2	BVDVIRSER(P)23-3	BVDVIRSER(P)23-4	BVDVIRSER(P)23-1
SP4	BVDVIRSER(P)23-4	BVDVIRSER(P)23-1	BVDVIRSER(P)23-3	BVDVIRSER(P)23-5
SN1	BVDVIRSER(P)23-1	BVDVIRSER(P)23-5	BVDVIRSER(P)23-5	BVDVIRSER(P)23-3

Sample content: PT2023 BVDVIR_	97514	97534	97540
SP1	BVDVIRSER(P)23-4	BVDVIRSER(P)23-4	BVDVIRSER(P)23-1
SP2	BVDVIRSER(P)23-3	BVDVIRSER(P)23-3	BVDVIRSER(P)23-3
SP3	BVDVIRSER(P)23-5	BVDVIRSER(P)23-2	BVDVIRSER(P)23-4
SP4	BVDVIRSER(P)23-2	BVDVIRSER(P)23-5	BVDVIRSER(P)23-2
SN1	BVDVIRSER(P)23-1	BVDVIRSER(P)23-1	BVDVIRSER(P)23-5

## 3.4 Virology (blood - ELISA)

### 3.4.1 THE PARTICIPANTS

Four laboratories participated in the proficiency test of BVD virology on blood (ELISA). The names of the participating laboratories are:

- Sciensano, department of Veterinary Virology
- ARSIA
- Dierengezondheidszorg Vlaanderen (DGZ)
- LAVETAN

### 3.4.2 THE SAMPLES

The samples (frozen blood) were prepared by the National Reference Laboratory (NRL), Service of VIRENBEE.

Information about the **origin** of the samples:

- Sample BP1: 16,05,2014 cat vanderperre 1
- Sample BP2: 12092014 cat dox1
- Sample BP3: 25022012CATBVDCER7
- Sample BP4: PONCIN SOLANGE
- Sample BN1: 24.08.2012 CATZELE 17

All samples are field samples stored frozen before aliquotation and transport.

### 3.4.3 HOMOGENEITY

The homogeneity of the samples was tested by the NRL on three aliquots (three blood samples, each 250 µL) of each sample using ELISA before and after the PT. The NRL obtained each time the same qualitative result. Therefore, the samples were considered as homogeneous.

### 3.4.4 TARGET VALUES

The target values were determined by the NRL based on the homogeneity tests. The panel consisted of five different samples. However, positive sample BP2 was replicated two times. Additionally, negative sample BN1 was repeated two times. Therefore, the panel included seven samples in total.

Sample content	Repetition	Expected result
PT2023BVDAgVIR_BP1	1	POS
PT2023BVDAgVIR_BP2	2	POS
PT2023BVDAgVIR_BP3	1	POS
PT2023BVDAgVIR_BP4	1	POS
PT2023BVDAgVIR_BN1	2	NEG

(POS = positive; NEG = negative)

### 3.4.5 STABILITY

The stability was determined by comparison of the pre-proficiency test results with the results obtained by the NRL during and after the proficiency test. The samples were considered as stable.

### 3.4.6 RANDOMISATION AND PANEL COMPOSITION

Since a specific number has been assigned to each laboratory, the randomisation has been performed as follows:

Sample content: PT2023 BVDAg VIR_	97505	97507	97508	97509
BP1	BVDVIR BLOOD(E)23-1	BVDVIR BLOOD(E)23-5	BVDVIR BLOOD(E)23-2	BVDVIR BLOOD(E)23-1
BP2 (1)	BVDVIR BLOOD(E)23-5	BVDVIR BLOOD(E)23-1	BVDVIR BLOOD(E)23-1	BVDVIR BLOOD(E)23-2
BP2 (2)	BVDVIR BLOOD(E)23-6	BVDVIR BLOOD(E)23-4	BVDVIR BLOOD(E)23-6	BVDVIR BLOOD(E)23-5
BP3	BVDVIR BLOOD(E)23-7	BVDVIR BLOOD(E)23-6	BVDVIR BLOOD(E)23-3	BVDVIR BLOOD(E)23-7
BP4	BVDVIR BLOOD(E)23-2	BVDVIR BLOOD(E)23-7	BVDVIR BLOOD(E)23-7	BVDVIR BLOOD(E)23-6
BN1 (1)	BVDVIR BLOOD(E)23-3	BVDVIR BLOOD(E)23-2	BVDVIR BLOOD(E)23-4	BVDVIR BLOOD(E)23-3
BN1 (2)	BVDVIR BLOOD(E)23-4	BVDVIR BLOOD(E)23-3	BVDVIR BLOOD(E)23-5	BVDVIR BLOOD(E)23-4

### 3.4.7 THRESHOLD FOR QUALIFICATION

Following the procedure, a participating laboratory is only qualified if the level of agreement for the seven reference samples is 100%.

## 3.5 Virology (blood - RT-qPCR)

### 3.5.1 THE PARTICIPANTS

Seven laboratories participated in the proficiency test of BVD virology on blood (RT-qPCR). The names of the participating laboratories are:

- Sciensano, department of Veterinary Virology
- ARSIA
- Dierengezondheidszorg Vlaanderen (DGZ)
- Biosellal
- Laboratoire de Médecine Vétérinaire de l'Etat (LMVE)
- LSI-ThermoFisher Scientific (France)
- Poulpharm

### 3.5.2 THE SAMPLES

The samples (frozen blood) were prepared by the National Reference Laboratory (NRL), Service of VIRENBEE, Sciensano.

Information about the **origin** of the samples:

- Sample BP1: 04.12.12 CATBRAEM 0032
- Sample BP2: 01102012CATRendac1 - DIL 1/50
- Sample BP3: PONCIN SOLANGE
- Sample BP4: 12092014 cat dox1
- Sample BN1: 24.08.2012 CATZELE20

All samples are field samples stored frozen before aliquotation and transport.

### 3.5.3 HOMOGENEITY

The homogeneity of the samples was tested by the NRL on three aliquots (three blood samples, each 500 µL) of each sample using RT-PCR before and after the PT. The NRL obtained each time the same qualitative result. Therefore, the samples were considered as homogeneous.

### 3.5.4 TARGET VALUES

The target values were determined by the NRL based on the homogeneity tests. The panel consisted of five different samples. No repetitions were included.

Sample content	Repetition	Expected result
PT2023BVDVIR_BP1	1	POS
PT2023BVDVIR_BP2	1	POS
PT2023BVDVIR_BP3	1	POS
PT2023BVDVIR_BP4	1	POS
PT2023BVDVIR_BN1	1	NEG

(POS = positive; NEG = negative)

### 3.5.5 STABILITY

The stability was determined by comparison of the pre-proficiency test results with the results obtained by the NRL during and after the proficiency test. The samples were considered as stable.

### 3.5.6 RANDOMISATION AND PANEL COMPOSITION

Since a specific number has been assigned to each laboratory, the randomisation has been performed as follows:

Sample content: PT2023 BVDVIR_	97505	97507	97508	97514
BP1	BVDVIR BLOOD(P)23-1	BVDVIR BLOOD(P)23-5	BVDVIR BLOOD(P)23-2	BVDVIR BLOOD(P)23-4
BP2	BVDVIR BLOOD(P)23-4	BVDVIR BLOOD(P)23-2	BVDVIR BLOOD(P)23-4	BVDVIR BLOOD(P)23-1
BP3	BVDVIR BLOOD(P)23-5	BVDVIR BLOOD(P)23-4	BVDVIR BLOOD(P)23-3	BVDVIR BLOOD(P)23-2
BP4	BVDVIR BLOOD(P)23-2	BVDVIR BLOOD(P)23-1	BVDVIR BLOOD(P)23-1	BVDVIR BLOOD(P)23-3
BN1	BVDVIR BLOOD(P)23-3	BVDVIR BLOOD(P)23-3	BVDVIR BLOOD(P)23-5	BVDVIR BLOOD(P)23-5

Sample content: PT2023 BVDVIR_	97516	97534	97540
BP1	BVDVIR BLOOD(P)23-1	BVDVIR BLOOD(P)23-3	BVDVIR BLOOD(P)23-3
BP2	BVDVIR BLOOD(P)23-5	BVDVIR BLOOD(P)23-2	BVDVIR BLOOD(P)23-4
BP3	BVDVIR BLOOD(P)23-2	BVDVIR BLOOD(P)23-5	BVDVIR BLOOD(P)23-5
BP4	BVDVIR BLOOD(P)23-4	BVDVIR BLOOD(P)23-1	BVDVIR BLOOD(P)23-2
BN1	BVDVIR BLOOD(P)23-3	BVDVIR BLOOD(P)23-4	BVDVIR BLOOD(P)23-1

### 3.5.7 THRESHOLD FOR QUALIFICATION

Following the procedure, a participating laboratory is only qualified if the level of agreement for the five reference samples is 100%.

## 3.6 Virology (ear notch - ELISA)

### 3.6.1 THE PARTICIPANTS

Five laboratories participated in the proficiency test of BVD virology on ear notch (RT-qPCR). The names of the participating laboratories are:

- Sciensano, department of Veterinary Virology
- ARSIA
- Dierengezondheidszorg Vlaanderen (DGZ)
- LAVETAN
- ANSES Unité Pathologie et Bien-être des ruminants (PBER)-Site de Niort

### 3.6.2 THE SAMPLES

The samples (frozen ear notch) were prepared by the National Reference Laboratory (NRL), Service of VIRENBEE, Sciensano.

Information about the **origin** of the samples:

- Sample EP1: 12.09.2014CATDOX1
- Sample EP2: 16.05.2014CATVAN DEN PERRE
- Sample EP3: BE9663667793
- Sample EN1: be61434463
- Sample EN2: be471474094

All samples are ear samples collected from IPI and non-IPI animals in Belgium, kept frozen at Sciensano.

### 3.6.3 HOMOGENEITY

The homogeneity of the samples was tested by the NRL on three aliquots (three ear notch samples) of each sample using ELISA before and after the PT. The NRL obtained each time the same qualitative result. Therefore, the samples were considered as homogeneous.

### 3.6.4 TARGET VALUES

The target values were determined by the NRL based on the homogeneity tests. The panel consisted of five different samples. No repetitions were included.

Sample content	Repetition	Expected result
PT2023BVDAg_EP1	1	POS
PT2023BVDAg_EP2	1	POS
PT2023BVDAg_EP3	1	POS
PT2023BVDAg_EN1	1	NEG
PT2023BVDAg_EN2	1	NEG

(POS = positive; NEG = negative)

### 3.6.5 STABILITY

The stability was determined by comparison of the pre-proficiency test results with the results obtained by the NRL during and after the proficiency test. The samples were considered as stable.

### 3.6.6 RANDOMISATION AND PANEL COMPOSITION

Since a specific number has been assigned to each laboratory, the randomisation has been performed as follows:

Sample content: PT 2023BVDAg_	97505	97507	97508	97509	97513
EP1	BVDVIR EN(E)23-2	BVDVIR EN(E)23-1	BVDVIR EN(E)23-4	BVDVIR EN(E)23-4	BVDVIR EN(E)23-5
EP2	BVDVIR EN(E)23-4	BVDVIR EN(E)23-5	BVDVIR EN(E)23-2	BVDVIR EN(E)23-5	BVDVIR EN(E)23-1
EP3	BVDVIR EN(E)23-1	BVDVIR EN(E)23-2	BVDVIR EN(E)23-3	BVDVIR EN(E)23-2	BVDVIR EN(E)23-3
EN1	BVDVIR EN(E)23-3	BVDVIR EN(E)23-3	BVDVIR EN(E)23-5	BVDVIR EN(E)23-1	BVDVIR EN(E)23-2
EN2	BVDVIR EN(E)23-5	BVDVIR EN(E)23-4	BVDVIR EN(E)23-1	BVDVIR EN(E)23-3	BVDVIR EN(E)23-4

### 3.6.7 THRESHOLD FOR QUALIFICATION

Following the procedure, a participating laboratory is only qualified if the level of agreement for the five reference samples is 100%.

## 3.7 Virology (ear notch - RT-qPCR)

### 3.7.1 THE PARTICIPANTS

Nine laboratories participated in the proficiency test of BVD virology on ear notch (RT-qPCR). The names of the participating laboratories are:

- Sciensano, department of Veterinary Virology
- ARSIA
- Dierengezondheidszorg Vlaanderen (DGZ)
- ANSES Unité Pathologie et Bien-être des ruminants (PBER)-Site de Niort
- Biosellal
- Laboratoire de Médecine Vétérinaire de l'Etat (LMVE)
- IDVET
- LSI-ThermoFisher Scientific (France)
- Poulpharm



### 3.7.2 THE SAMPLES

The samples (frozen ear notch) were prepared by the National Reference Laboratory (NRL), Service of VIRENBEE, Sciensano.

Information about the **origin** of the samples:

- Sample EP1: TO-16-042472
- Sample EP2: be 456129869
- Sample EP3: 12.09.2014 Catdox1
- Sample EN1: BE512670801
- Sample EN2: BE815561529

All samples are ear samples collected from IPI and non-IPI animals in Belgium, kept frozen at Sciensano.

### 3.7.3 HOMOGENEITY

The homogeneity of the samples was tested by the NRL on three aliquots (three ear notch samples) of each sample using RT-PCR before and after the PT. The NRL obtained each time the same qualitative result. Therefore, the samples were considered as homogeneous.

### 3.7.4 TARGET VALUES

The target values were determined by the NRL based on the homogeneity tests. The panel consisted of five different samples. No repetitions were included.

Sample content	Repetition	Expected result
PT2023BVDVIR_EP1	1	POS
PT2023BVDVIR_EP2	1	POS
PT2023BVDVIR_EP3	1	POS
PT2023BVDVIR_EN1	1	NEG
PT2023BVDVIR_EN2	1	NEG

(POS = positive; NEG = negative)

### 3.7.5 STABILITY

The stability was determined by comparison of the pre-proficiency test results with the results obtained by the NRL during and after the proficiency test. The samples were considered as stable.

### 3.7.6 RANDOMISATION AND PANEL COMPOSITION

Since a specific number has been assigned to each laboratory, the randomisation has been performed as follows:

Sample content: PT2023 BVDVIR_	97505	97507	97508	97513	97514	97516
EP1	BVDVIR EN(P)23-1	BVDVIR EN(P)23-2	BVDVIR EN(P)23-3	BVDVIR EN(P)23-3	BVDVIR EN(P)23-5	BVDVIR EN(P)23-1
EP2	BVDVIR EN(P)23-2	BVDVIR EN(P)23-4	BVDVIR EN(P)23-2	BVDVIR EN(P)23-4	BVDVIR EN(P)23-1	BVDVIR EN(P)23-2
EP3	BVDVIR EN(P)23-3	BVDVIR EN(P)23-3	BVDVIR EN(P)23-4	BVDVIR EN(P)23-5	BVDVIR EN(P)23-2	BVDVIR EN(P)23-5
EN1	BVDVIR EN(P)23-4	BVDVIR EN(P)23-5	BVDVIR EN(P)23-1	BVDVIR EN(P)23-1	BVDVIR EN(P)23-4	BVDVIR EN(P)23-3
EN2	BVDVIR EN(P)23-5	BVDVIR EN(P)23-1	BVDVIR EN(P)23-5	BVDVIR EN(P)23-2	BVDVIR EN(P)23-3	BVDVIR EN(P)23-4

Sample content: PT2023 BVDVIR_	97522	97534	97540
EP1	BVDVIREN(P)23-4	BVDVIREN(P)23-3	BVDVIREN(P)23-2
EP2	BVDVIREN(P)23-2	BVDVIREN(P)23-5	BVDVIREN(P)23-5
EP3	BVDVIREN(P)23-5	BVDVIREN(P)23-1	BVDVIREN(P)23-1
EN1	BVDVIREN(P)23-1	BVDVIREN(P)23-4	BVDVIREN(P)23-4
EN2	BVDVIREN(P)23-3	BVDVIREN(P)23-2	BVDVIREN(P)23-3

### 3.7.7 THRESHOLD FOR QUALIFICATION

Following the procedure, a participating laboratory is only qualified if the level of agreement for the five reference samples is 100%.

## 4 TIMELINE

Transfer of the samples from NRL to QL: 29/09/2023

Randomization of the samples by QL: 05/10/2023

Sending samples to participants: 09/10/2023

Deadline for submitting the results: 27/10/2023

Definitive individual report: 07/12/2023

## 5 RESULTS

### 5.1 Serology (serum - ELISA)

#### 5.1.1 RESULTS PER SAMPLE

The panel consisted of four different samples. However, positive sample PS1 and negative sample NS1 were replicated three times. Additionally, positive samples PS2 and PS3 were repeated two times. Therefore, the panel included ten samples in total.

Two labs had chosen to test more than one method on the same samples, implying that one lab submitted two datasets and one lab submitted three datasets. These additional results are included in the tables below.

Sample ID	Status	Number of repetitions (total results)	Observed result
PS1	POS	3 (33)	33 POS
PS2	POS	2 (22)	22 POS
PS3	POS	2 (22)	22 POS
NS1	NEG	3 (33)	33 NEG

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

#### 5.1.2 USED ELISA PROTOCOL/KIT

In the table below, the ELISA protocols/kits used are listed along with the number of datasets and their achieved score.

Method	Name producer	Name kit	N	NR	NCR	%
ELISA Competition	Bio-X Diagnostics	Monoscreen Ab ELISA BVD	4	40	40	100
ELISA Competition	ID.VET	ID screen BVD p80 antibody competition	5	50	50	100
ELISA Indirect	IDEXX	BVDV Total Ab X3	2	20	20	100
<b>TOTAL</b>			<b>11</b>	<b>110</b>	<b>110</b>	<b>100</b>

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

#### 5.1.3 CONCLUSION

In 2023, eight laboratories participated in the proficiency test BVD serology (serum - ELISA) organized by Sciansano. According to the procedure currently in force, the performance of a participating laboratory is satisfactory if at least 90% of the results provided by the laboratory are in agreement with the status of the reference samples assigned by the NRL of the Scientific Directorate Infectious Diseases in Animals of Sciansano. All laboratories succeeded in achieving the maximum score (100%) for this test.

## 5.2 Virology (serum - ELISA)

### 5.2.1 RESULTS PER SAMPLE

The panel consisted of four different samples. However, positive sample SP1 was replicated two times. Additionally, negative samples SN1 and SN2 were repeated two times. Therefore, the panel included seven samples in total.

Sample ID	Status	Number of repetitions (total results)	Observed result
SP1	POS	2 (10)	10 POS
SP2	POS	1 (5)	5 POS
SN1	NEG	2 (10)	10 NEG
SN2	NEG	2 (10)	10 NEG

(POS = positive; NEG = negative)

### 5.2.2 USED ELISA PROTOCOL/KIT

In the table below, the ELISA protocols/kits used are listed along with the number of laboratories that have used this protocol/kits with their achieved score.

Method	Name producer	Name kit	N	NR	NCR	%
ELISA Sandwich	IDEXX	BVDV Ag/Serum Plus Test	5	35	35	100
<b>TOTAL</b>			<b>5</b>	<b>35</b>	<b>35</b>	<b>100</b>

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

### 5.2.3 CONCLUSION

In 2023, five laboratories participated in the proficiency test BVD virology (serum - ELISA) organized by Sciensano. According to the procedure currently in force, the performance of a participating laboratory is satisfactory if all the results (100%) provided by the laboratory are in agreement with the status of the reference samples assigned by the NRL of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test.

## 5.3 Virology (serum – RT-qPCR)

### 5.3.1 RESULTS PER SAMPLE

The panel consisted of five different samples. No repetitions were included.

Sample ID	Status	Number of repetitions (total results)	Observed result
SP1	POS	1 (7)	7 POS
SP2	POS	1 (7)	7 POS
SP3	POS	1 (7)	7 POS
SP4	POS	1 (7)	7 POS
SN1	NEG	1 (7)	7 NEG

(POS = positive; NEG = negative)

### 5.3.2 USED RT-PCR PROTOCOL/KIT

In the table below, the RT-PCR protocols/kits used are listed along with the number of laboratories that have used this protocol/kit with their achieved score.

Manufacturer RT-qPCR protocol / kit	Name RT-qPCR protocol / kit	N	NR	NCR	%
Homemade	Homemade	1	5	5	100
Thermo Fisher Scientific	LSI VETMAX BVD4ALL	3	15	15	100
ADIAGENE	ADIAVET BVD real time	1	5	5	100
Biosellal	Bio-T kit® BVDV-BDV Universal	1	5	5	100
IDEXX	RealPCR BVDV RNA test	1	5	5	100
<b>TOTAL</b>		<b>7</b>	<b>35</b>	<b>35</b>	<b>100</b>

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

### 5.3.3 USED EXTRACTION PROTOCOL

In the table below, the extraction protocols/kits used are listed along with the number of laboratories that have used this protocol/kit with their achieved score.

Manufacturer extraction protocol / kit	Name extraction protocol / kit	N	NR	NCR	%
QIAGEN	Rneasy mini kit	1	5	5	100
Indical Bioscience	IndiMag Pathogen Kit	2	10	10	100
Bio-X ADIAGENE	ADIAMAG XL	1	5	5	100
Biosellal	BioExtract® Column	1	5	5	100
Thermo Fisher Scientific	MagMAX Core Nucleic Acid purification kit	1	5	5	100
QIAGEN	QIAamp Viral RNA Mini Kit	1	5	5	100
<b>TOTAL</b>		<b>7</b>	<b>35</b>	<b>35</b>	<b>100</b>

### 5.3.4 CONCLUSION

In 2023, seven laboratories participated in the proficiency test BVD virology (serum – RT-qPCR) organized by Sciensano. According to the procedure currently in force, the performance of a participating laboratory is satisfactory if all the results (100%) provided by the laboratory are in agreement with the status of the reference samples assigned by the NRL of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test.

## 5.4 Virology (blood – ELISA)

### 5.4.1 RESULTS PER SAMPLE

The panel consisted of five different samples. However, positive sample BP2 was replicated two times. Additionally, negative sample BN1 was repeated two times. Therefore, the panel included seven samples in total.

Sample ID	Status	Number of repetitions (total results)	Observed result
BP1	POS	1 (7)	7 POS
BP2	POS	2 (14)	14 POS
BP3	POS	1 (7)	7 POS
BP4	POS	1 (7)	7 POS
BN1	NEG	2 (14)	14 NEG

(POS = positive; NEG = negative)

### 5.4.2 USED ELISA PROTOCOL/KIT

In the table below, the ELISA protocols/kits used are listed along with the number of laboratories that have used this protocol/kit with their achieved score.

Method	Name producer	Name kit	N	NR	NCR	%
ELISA Sandwich	IDEXX	BVDV Ag/Serum Plus Test	4	28	28	100
<b>TOTAL</b>			<b>4</b>	<b>28</b>	<b>28</b>	<b>100</b>

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

### 5.4.3 CONCLUSION

In 2023, four laboratories participated in the proficiency test BVD virology (blood - ELISA) organized by Sciensano. According to the procedure currently in force, the performance of a participating laboratory is satisfactory if all the results (100%) provided by the laboratory are in agreement with the status of the reference samples assigned by the NRL of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test.

## 5.5 Virology (blood - RT-qPCR)

### 5.5.1 RESULTS PER SAMPLE

The panel consisted of five different samples. No repetitions were included.

Sample ID	Status	Number of repetitions (total results)	Observed result
BP1	POS	1 (5)	5 POS
BP2	POS	1 (5)	5 POS
BP3	POS	1 (5)	5 POS
BP4	POS	1 (5)	5 POS
BN1	NEG	1 (5)	5 NEG

(POS = positive; NEG = negative)

### 5.5.2 USED RT-PCR PROTOCOL/KIT

In the table below, the RT-PCR protocols/kits used are listed along with the number of laboratories that have used this protocol/kit with their achieved score.

Manufacturer RT-qPCR protocol / kit	Name RT-qPCR protocol / kit	N	NR	NCR	%
Homemade	Homemade	1	5	5	100
Thermo Fisher Scientific	LSI VETMAX BVD4ALL	3	15	15	100
Applied Biosystems	VetMAX™BVDV4ALL kit	1	5	5	100
Biosellal	Bio-T kit® BVDV-BDV Universal	1	5	5	100
IDEXX	RealPCR BVDV RNA test	1	5	5	100
<b>TOTAL</b>		<b>6</b>	<b>30</b>	<b>30</b>	<b>100</b>

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



### 5.5.3 USED EXTRACTION PROTOCOL

In the table below, the extraction protocols/kits used are listed along with the number of laboratories that have used this protocol/kit with their achieved score.

Manufacturer extraction protocol / kit	Name extraction protocol / kit	N	NR	NCR	%
QIAGEN	Rneasy mini kit	1	5	5	100
Indical Bioscience	IndiMag Pathogen Kit	2	10	10	100
Biosellal	BioExtract® Column	1	5	5	100
Thermo Fisher Scientific	MagMAX Core Nucleic Acid Purification Kit	2	10	10	100
QIAGEN	QIAamp Viral RNA Mini Kit	1	5	5	100
<b>TOTAL</b>		<b>7</b>	<b>35</b>	<b>35</b>	<b>100</b>

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

### 5.5.4 CONCLUSION

In 2023, seven laboratories participated in the proficiency test BVD virology (blood – RT-qPCR) organized by Sciensano. According to the procedure currently in force, the performance of a participating laboratory is satisfactory if all the results (100%) provided by the laboratory are in agreement with the status of the reference samples assigned by the NRL of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test.

## 5.6 Virology (ear notch - ELISA)

### 5.6.1 RESULTS PER SAMPLE

The panel consisted of five different samples. No repetitions were included.

Sample ID	Status	Number of repetitions (total results)	Observed result
EP1	POS	1 (5)	5 POS
EP2	POS	1 (5)	5 POS
EP3	POS	1 (5)	5 POS
EN1	NEG	1 (5)	5 NEG
EN2	NEG	1 (5)	5 NEG

(POS = positive; NEG = negative)

### 5.6.2 USED ELISA PROTOCOL/KIT

In the table below, the ELISA protocols/kits used are listed along with the number of laboratories that have used this protocol/kit with their achieved score.

Method	Name producer	Name kit	N	NR	NCR	%
ELISA Sandwich	IDEXX	BVD Ag/serum plus test kit	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)

### 5.6.3 CONCLUSION

In 2023, five laboratories participated in the proficiency test BVD virology (ear notch - ELISA) organized by Sciensano. According to the procedure currently in force, the performance of a participating laboratory is satisfactory if all the results (100%) provided by the laboratory are in agreement with the status of the reference samples assigned by the NRL of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test.

## 5.7 Virology (ear notch - RT-qPCR)

### 5.7.1 RESULTS PER SAMPLE

The panel consisted of five different samples. No repetitions were included.

Sample ID	Status	Number of repetitions (total results)	Observed result
EP1	POS	1 (9)	9 POS
EP2	POS	1 (9)	9 POS
EP3	POS	1 (9)	9 POS
EN1	NEG	1 (9)	9 NEG
EN2	NEG	1 (9)	9 NEG

(POS = positive; NEG = negative)

### 5.7.2 USED RT-PCR PROTOCOL/KIT

In the table below, the RT-PCR protocols/kits used are listed along with the number of laboratories that have used this protocol/kit with their achieved score.

Manufacturer RT-qPCR protocol / kit	Name RT-qPCR protocol / kit	N	NR	NCR	%
Homemade	Homemade	1	5	5	100
Thermo Fisher Scientific	LSI VETMAX BVD4ALL	3	15	15	100
QIAGEN	BVD RT-PCR kit	1	5	5	100
BioX-Adiagene	Adiavet BVD RealTime	1	5	5	100
Biosellal	Bio-T kit® BVDV-BDV Universal	1	5	5	100
ID.VET	ID GENE® BVD/BD TRIPLEX	1	5	5	100
IDEXX	RealPCR BVDV RNA test	1	5	5	100
<b>TOTAL</b>		<b>9</b>	<b>45</b>	<b>45</b>	<b>100</b>

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

### 5.7.3 USED EXTRACTION PROTOCOL

In the table below, the extraction protocols/kits used are listed along with the number of laboratories that have used this protocol/kit with their achieved score.

Manufacturer extraction protocol / kit	Name extraction protocol / kit	N	NR	NCR	%
QIAGEN	RNeasy minikit	2	10	10	100
Indical Bioscience	IndiMag Pathogen Kit	1	5	5	100
IDVET	Direct Lysis Buffer.	1	5	5	100
Bio-X ADIAGENE	ADIAMAG XL	1	5	5	100
Biosellal	BioExtract® Column	1	5	5	100
Thermo Fisher Scientific	MagMAX Core Nucleic Acid Purification Kit	2	10	10	100
IDVET	ID Gene Easy Preparation of Ear notch samples 2.0	1	5	5	100
<b>TOTAL</b>		<b>9</b>	<b>45</b>	<b>45</b>	<b>100</b>

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

### 5.7.4 CONCLUSION

In 2023, nine laboratories participated in the proficiency test BVD virology (ear notch – RT-qPCR) organized by Sciensano. According to the procedure currently in force, the performance of a participating laboratory is satisfactory if all the results (100%) provided by the laboratory are in agreement with the status of the reference samples assigned by the NRL of the Scientific Directorate Infectious Diseases in Animals of Sciensano. All laboratories succeeded in achieving the maximum score (100%) for this test.

## 6 ANNEXES (NOT UNDER ACCREDITATION)

This quantitative data is not under BELAC-accreditation and is solely for the information of the laboratories.

### 6.1 Annex 1: Quantitative results

Boxplots are generated exclusively for the positive samples that exhibited repetitions within the panel. The boxplots, shown down below, were created by using the following software programme: [shiny.chemgrid.org/boxplotr/](https://shiny.chemgrid.org/boxplotr/).

#### 6.1.1 SEROLOGY SERUM (ELISA)

PT2023BVDAbSER\_PS1

Lab number	97505	97507	97508	97509	97513 (1)	97513 (2)	97513 (3)	97540	97544	97621 (1)	97621 (2)
Method (ELISA protocol /kit)	M <sub>1</sub>	M <sub>1</sub>	M <sub>1</sub>	M <sub>2</sub>	M <sub>2</sub>	M <sub>1</sub>	M <sub>3</sub>	M <sub>2</sub>	M <sub>3</sub>	M <sub>2</sub>	M <sub>2</sub>
S/P % (REP1)	71,08	76,25	63,64	70,98	25,00	68,00	46,27	35,05	61,53	26,00	36,00
S/P % (REP2)	70,63	76,83	68,45	71,07	25,00	69,00	45,14	32,98	37,87	28,00	39,00
S/P % (REP3)	70,65	73,52	66,13	71,60	22,00	70,00	53,25	32,35	43,41	29,00	40,00
Mean	70,79	75,53	66,07	71,22	24,00	69,00	48,22	33,46	47,60	49,00	38,33
SD	0,25	1,77	2,41	0,33	1,73	1,00	4,40	1,41	12,37	38,12	2,08
CV (%)	0,36	2,34	3,64	0,47	7,22	1,45	9,12	4,21	25,98	77,79	5,43

Numbers were rounded to two significant decimal place. (S/P = Signal-to-Positive ratio; REP = repetition; SD = standard deviation; CV = coefficient of variation; M<sub>1</sub> = Bio-X Diagnostics - Monoscreen Ab ELISA BVD; M<sub>2</sub> = ID.VET - ID screen BVD p80 antibody competition; M<sub>3</sub> = IDEXX- BVDV Total Ab X3).

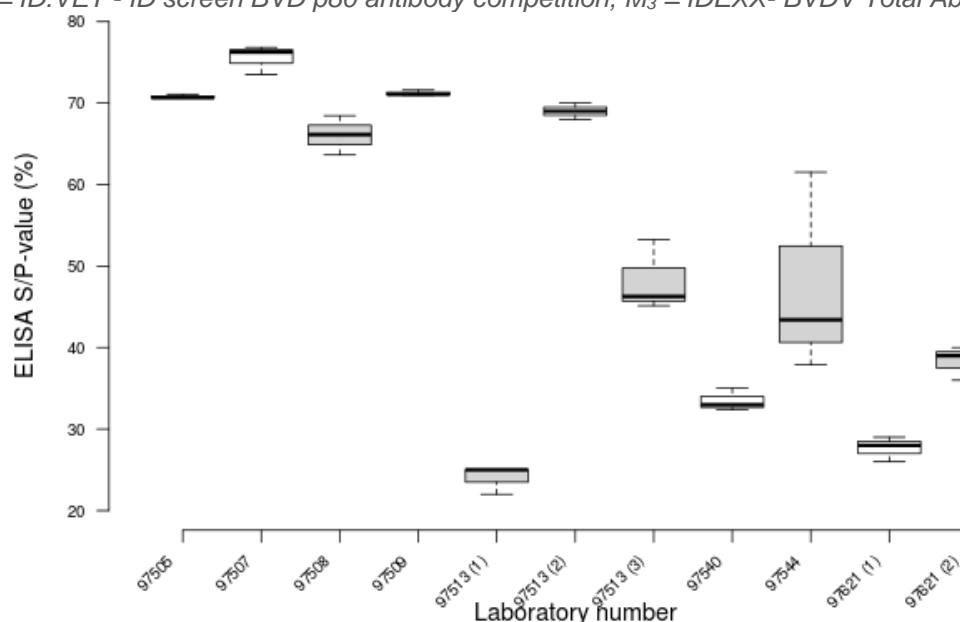


Figure 1. Distribution of the S/P-values (%) (box-plots) per laboratory.

Lab number	97505	97507	97508	97509	97513 (1)	97513 (2)	97513 (3)	97540	97544	97621 (1)	97621 (2)
Method (ELISA protocol /kit)	M <sub>1</sub>	M <sub>1</sub>	M <sub>1</sub>	M <sub>2</sub>	M <sub>2</sub>	M <sub>1</sub>	M <sub>3</sub>	M <sub>2</sub>	M <sub>3</sub>	M <sub>2</sub>	M <sub>2</sub>
S/P % (REP1)	94,38	92,48	93,14	91,60	7,00	94,00	112,77	7,89	253,44	7,00	7,00
S/P % (REP2)	94,39	92,15	92,78	92,36	7,00	93,00	107,79	7,86	258,38	7,00	7,00
Mean	94,39	92,32	92,96	91,98	7,00	93,50	110,28	7,87	255,91	18,00	7,00
SD	0,0071	0,23	0,25	0,54	0,00	0,71	3,52	0,016	3,49	15,56	0,00
CV (%)	0,0075	0,25	0,27	0,58	0,00	0,76	3,19	0,20	1,381	86,42	0,00

Numbers were rounded to two significant decimal place. (S/P = Signal-to-Positive ratio; REP = repetition; SD = standard deviation; CV = coefficient of variation; M<sub>1</sub> = Bio-X Diagnostics - Monoscreen Ab ELISA BVD; M<sub>2</sub> = ID.VET - ID screen BVD p80 antibody competition; M<sub>3</sub> = IDEXX- BVDV Total Ab X3).

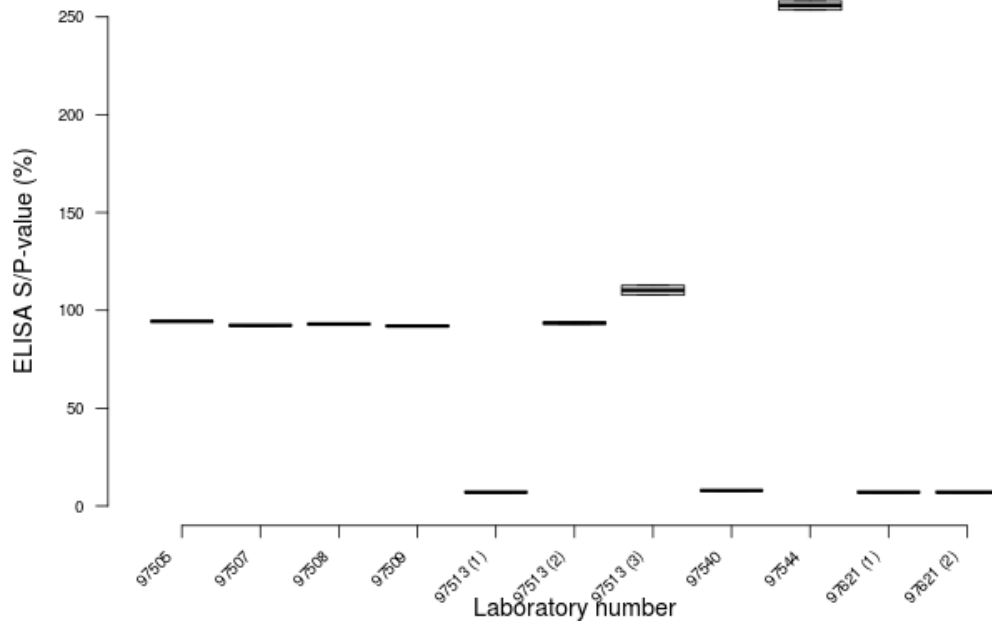


Figure 2. Distribution of the S/P-values (%) (box-plots) per laboratory.

Lab number	97505	97507	97508	97509	97513 (1)	97513 (2)	97513 (3)	97540	97544	97621 (1)	97621 (2)
Method (ELISA protocol /kit)	M <sub>1</sub>	M <sub>1</sub>	M <sub>1</sub>	M <sub>2</sub>	M <sub>2</sub>	M <sub>1</sub>	M <sub>3</sub>	M <sub>2</sub>	M <sub>3</sub>	M <sub>2</sub>	M <sub>2</sub>
S/P % (REP1)	72,78	82,39	76,16	86,84	12,00	81,00	109,72	15,04	199,25	13,00	13,00
S/P % (REP2)	80,47	82,92	74,91	84,98	11,00	79,00	106,35	15,35	202,10	14,00	17,00
Mean	76,63	82,66	75,54	85,91	11,50	80,00	108,03	15,19	200,68	13,50	15,00
SD	5,44	0,37	0,88	1,31	0,71	1,41	2,39	0,22	2,01	0,07	2,83
CV (%)	7,10	0,45	1,17	1,53	6,15	1,77	2,21	1,45	1,02	5,24	18,86

Numbers were rounded to two significant decimal place. (S/P = Signal-to-Positive ratio; REP = repetition; SD = standard deviation; CV = coefficient of variation; M<sub>1</sub> = Bio-X Diagnostics - Monoscreen Ab ELISA BVD; M<sub>2</sub> = ID.VET - ID screen BVD p80 antibody competition; M<sub>3</sub> = IDEXX- BVDV Total Ab X3).

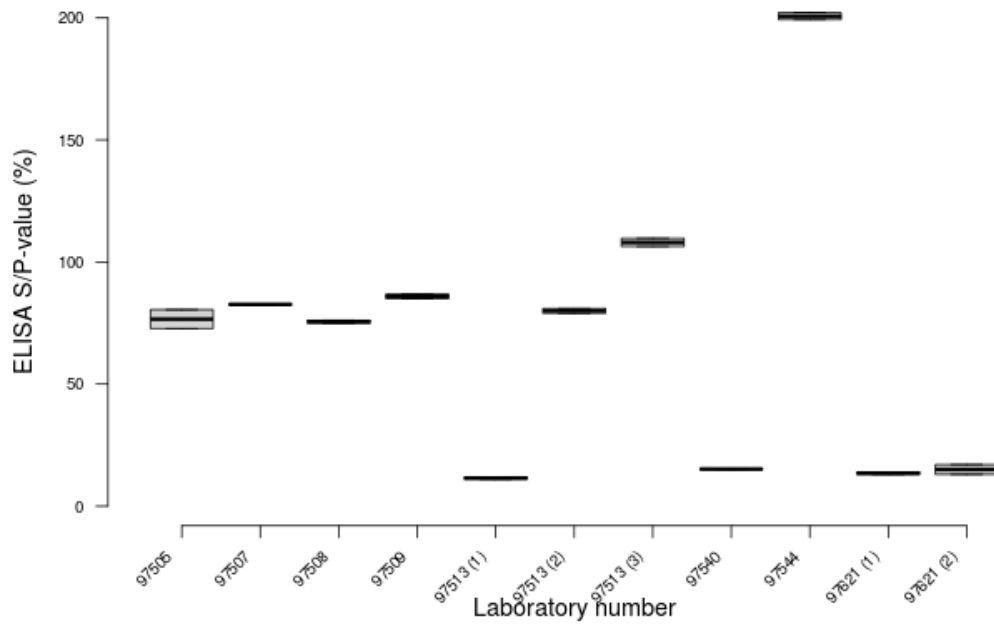


Figure 3. Distribution of the S/P-values (%) (box-plots) per laboratory.

## 6.1.2 VIROLOGY SERUM (ELISA)

PT2023BVDAgVIR\_SP1

Lab number	97505	97507	97508	97509	97513
Method (ELISA protocol/kit)	IDEXX - BVDV Ag/Serum Plus Test				
S/P % (REP1)	5,84	3,23	3,56	3,16	3,77
S/P % (REP2)	4,98	3,22	3,45	3,20	3,92
Mean	5,41	3,23	3,51	3,18	3,85
SD	0,61	0,0071	0,078	0,034	0,11
CV (%)	11,24	0,22	2,22	1,067	2,76

Numbers were rounded to two significant decimal place. (S/P = Signal-to-Positive ratio; REP = repetition; SD = standard deviation; CV = coefficient of variation).

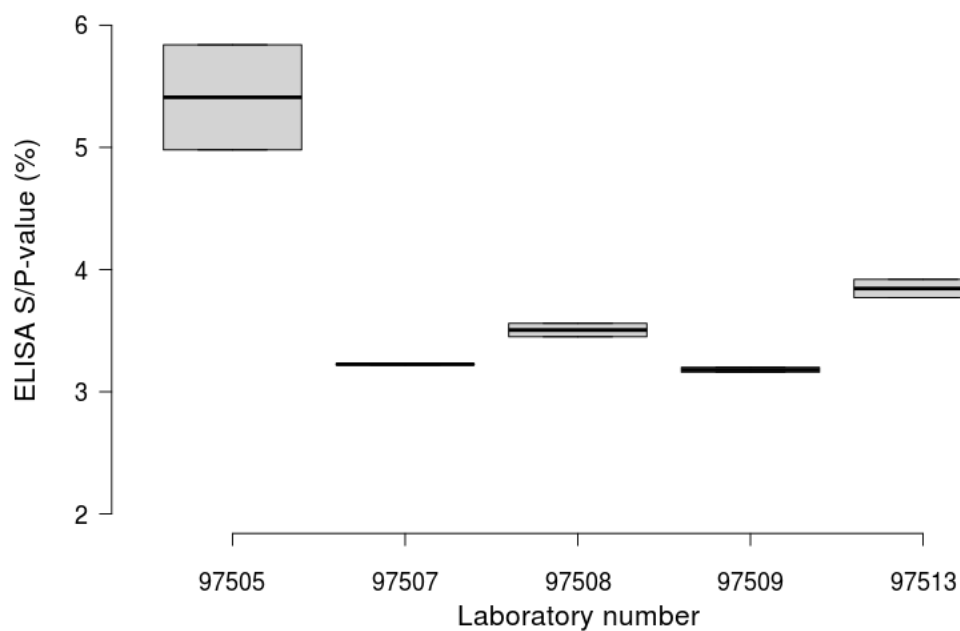


Figure 4. Distribution of the S/P-values (%) (box-plots) per laboratory.



### 6.1.3 VIROLOGY BLOOD (ELISA)

PT2023BVDAgVIR\_BP2

Lab number	97505	97507	97508	97509
Method (ELISA protocol/kit)	IDEXX - BVDV Ag/Serum Plus Test			
S/P % (REP1)	1,15	1,18	1,27	0,81
S/P % (REP2)	1,14	1,10	1,20	0,96
Mean	1,15	1,14	1,24	0,89
SD	0,0071	0,057	0,050	0,11
CV (%)	0,62	4,96	4,0079	11,96

Numbers were rounded to two significant decimal place. (S/P = Signal-to-Positive ratio; REP = repetition; SD = standard deviation; CV = coefficient of variation).

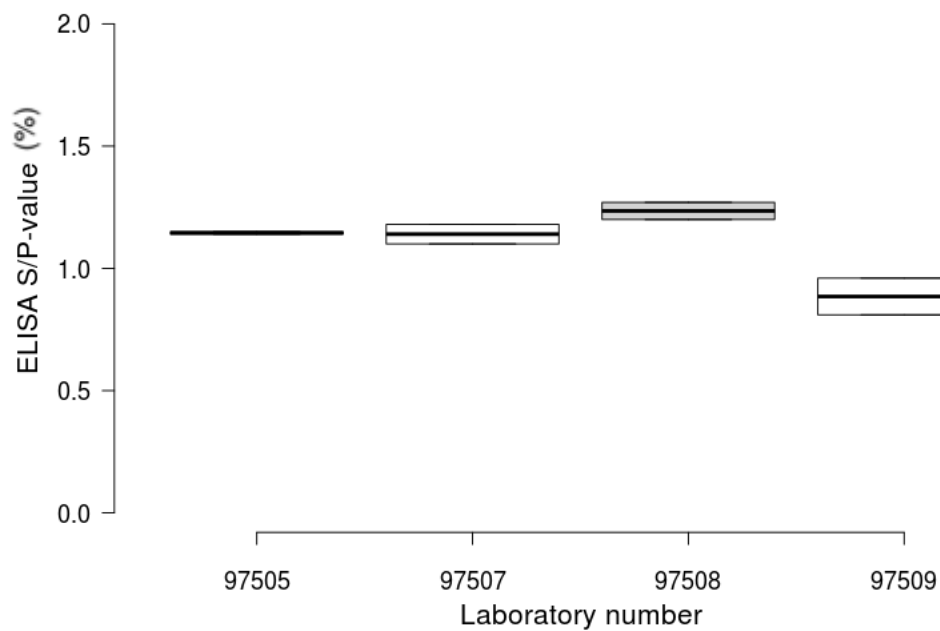


Figure 5. Distribution of the S/P-values (%) (box-plots) per laboratory.

## 6.2 Annex 2: Additional information

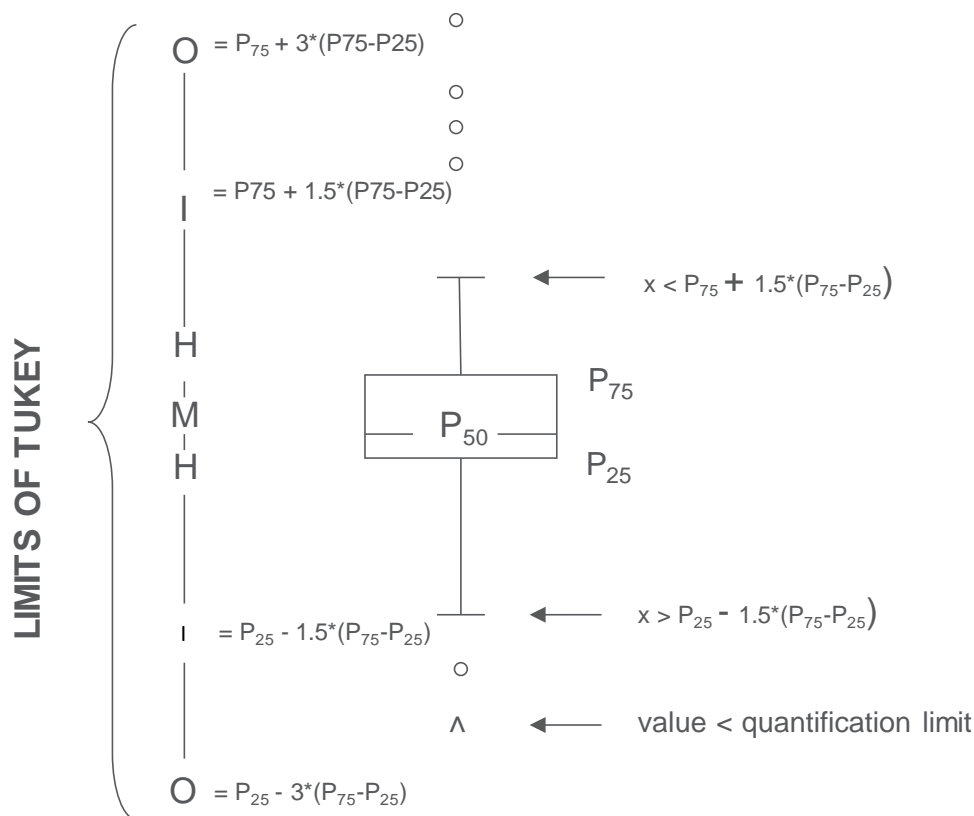
The **calendar** for Proficiency Testing in Veterinary diagnosis is available on our website:

- NL: <https://www.sciensano.be/fr/biblio/eke-kalender-2023>
- FR: <https://www.sciensano.be/en/biblio/calendrier-eeq-2023>
- EN: <https://www.sciensano.be/en/biblio/eqa-calendar-2023>

### Graphical representation

Besides the tables with the results a "Box and whisker" plot is added. It contains the following elements for the methods with at least 3 participants:

- a rectangle ranging from percentile 25 ( $P_{25}$ ) to percentile 75 ( $P_{75}$ )
- a central line representing the median of the results ( $P_{50}$ )
- a lower limit showing the smallest value  $x > P_{25} - 1.5 * (P_{75} - P_{25})$
- an upper limit representing the largest value  $x < P_{75} + 1.5 * (P_{75} - P_{25})$
- all points outside this interval are represented by a dot.



**Corresponding limits in case of normal distribution**

END

© Sciensano Brussels 2024.

This report may not be reproduced, published or distributed without the consent of Sciensano. The laboratories individual results are confidential. They are not passed on by Sciensano to third parties. Nevertheless, the results of FASFC licensed laboratories are transferred to FASFC.