

**BIOLOGICAL HEALTH RISKS
QUALITY OF LABORATORIES**

COMITEE OF EXPERTS

**EXTERNAL QUALITY ASSESSMENT
IN VETERINARY DIAGNOSIS**

DEFINITIVE GLOBAL REPORT

VETERINARY MEDECINE

**PORCINE REPRODUCTIVE AND RESPIRATORY SYNDROME
(PRRS)**

SURVEY 2022/1

Sciensano/PT VET PRRS/1-E/CV

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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 PT was dedicated to detect the agent of Porcine Reproductive and Respiratory Syndrome or PRRS (PRRS virus) by ELISA (Ab) in serum.

Initially, a PT virology was also 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.

3 MATERIALS AND METHODS

3.1 Serology on serum

3.1.1 THE PARTICIPANTS

Five laboratories participated in the proficiency test of PRRS Serology on serum. The names of the participating laboratories are:

- Sciensano
- ARSIA
- DGZ
- LAVETAN
- Poulpharm

3.1.2 THE SAMPLES

The samples were prepared by the National Reference Laboratory (NRL), service of Viral reemerging enzootic and BEE diseases, Infectious diseases in animals Directorate, Sciensano.

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

- PT2022PRRSSERSERUMNS1 and NS2 are two serum samples collected from two domestic pigs originated from a PRRS-free herd located in Flanders.
- PT2022PRRSSERSERUMNS3 is a serum sample collected from one domestic pig originated from a PRRS-free herd located in Wallonia.
- PT2022PRRSSERSERUMNS4 is a serum sample collected from one domestic pig originated from a PRRS-free Germinal center.
- PT2022PRRSSERSERUMPS5 is an undiluted serum collected from one domestic pig originated from a conventional herd and collected 23 days after experimental challenge with PRRSV (Flanders/13 strain).

Information about the **preparation** of the samples:

- PT2022PRRSERPS1, PS2, PS3 and PS4 are dilution 1/10, 1/8, 1/5 and 1/4 of the PT2022PRRSSERSERUMPS5 in a PRRSV-negative serum collected from a naïve animal.
- PT2022PRRSSERSERUMPS6 is an undiluted serum collected from one domestic pig originated from a PRRSV-free herd and collected at 14 days after experimental vaccination with Porcilis.
- PT2022PRRSSERSERUMPS7 is an undiluted serum collected from one domestic pig originated from a PRRSV-free herd and collected at 14 days after experimental vaccination with Ingelvac.
- PT2022PRRSSERSERUMPS8 is an undiluted serum collected from one domestic pig originated from a PRRSV-free herd and collected at 21 days after experimental vaccination with Unistrain.
- PT2022PRRSSERSERUMPS9 is a mix of sera from two domestic pigs originated from a PRRSV-free herd and collected at 21 days after vaccination (one animal with Suvaxyn / one animal with Ingelvac NA).
- PT2022PRRSSERSERUMPS10 and PS11 are serum samples collected from two domestic pigs from conventional herd vaccinated two times with PRRSFLEX (21 days apart) and experimentally challenge with PRRSV (Flanders/13 strain) 3 weeks after the second vaccination. The serum samples were collected 23 days after challenge.
- PT2022PRRSSERSERUMPS12 is serum sample collected from one domestic pig from conventional herd vaccinated one times with PRRSFLEX and experimentally challenge with PRRSV (Flanders/13 strain) 6 weeks after the vaccination. The serum samples was collected 23 days after challenge.

3.1.3 HOMOGENEITY

The homogeneity of the samples was tested by the NRL on 10 aliquots (100 µl) of each sample using the indirect ELISA method before the PT. 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 12 positive and 6 negative samples:

Sample ID	Status
PT2022PRRSSERSERUMPS1	POS
PT2022PRRSSERSERUMPS2	POS
PT2022PRRSSERSERUMPS3	POS
PT2022PRRSSERSERUMPS4	POS
PT2022PRRSSERSERUMPS5	POS
PT2022PRRSSERSERUMPS6	POS
PT2022PRRSSERSERUMPS7	POS
PT2022PRRSSERSERUMPS8	POS
PT2022PRRSSERSERUMPS9	POS
PT2022PRRSSERSERUMPS10	POS
PT2022PRRSSERSERUMPS11	POS
PT2022PRRSSERSERUMPS12	POS
PT2022PRRSSERSERUMNS1	NEG
PT2022PRRSSERSERUMNS2	NEG
PT2022PRRSSERSERUMNS3	NEG
PT2022PRRSSERSERUMNS4	NEG

(POS = positive; NEG = negative)

3.1.5 STABILITY

The samples were tested before and after the survey. The results were compared and 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 ID	97505	97507	97508	97509	97540
PRRSSERSERUM22-1	PS2	PS9	NS2	NS1	PS8
PRRSSERSERUM22-2	PS3	PS7	NS4	PS7	PS11
PRRSSERSERUM22-3	PS11	PS4	PS11	PS2	PS10
PRRSSERSERUM22-4	PS5	NS1	PS3	PS8	NS2
PRRSSERSERUM22-5	NS3	PS10	PS8	PS1	NS1
PRRSSERSERUM22-6	NS1	NS1	PS10	NS3	NS2
PRRSSERSERUM22-7	NS2	NS2	PS12	PS10	PS6
PRRSSERSERUM22-8	PS9	NS2	PS6	PS11	PS12
PRRSSERSERUM22-9	PS8	PS8	PS5	PS4	PS4
PRRSSERSERUM22-10	PS7	PS12	NS2	PS6	PS9
PRRSSERSERUM22-11	PS4	NS3	NS1	NS4	PS3
PRRSSERSERUM22-12	PS6	PS6	PS1	NS1	NS1
PRRSSERSERUM22-13	NS1	PS5	PS9	PS9	NS4
PRRSSERSERUM22-14	PS12	PS1	NS1	PS12	PS2
PRRSSERSERUM22-15	NS4	PS3	PS7	PS3	PS1
PRRSSERSERUM22-16	PS1	PS2	PS2	NS2	NS3
PRRSSERSERUM22-17	PS10	PS11	NS3	PS5	PS7
PRRSSERSERUM22-18	NS2	NS4	PS4	NS2	PS5

4 SURVEY TIMELINE

Transfer of the samples from NRL to QL: 22/02/2022

Randomization of the samples by QL: 24/02/2022

Sending samples (cooled at 4°C) to participants: 28/02/2022

Deadline for submitting the results: 28/03/2022

Preliminary report: 01/04/2022

5 RESULTS

5.1 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.

5.1.1 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)

5.1.2 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
TOTAL	7	126	126	100

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

5.1.3 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.

6 ANNEXES (NOT UNDER ACCREDITATION)

The boxplots, shown down below, were created by using the following software programme:
shiny.chemgrid.org/boxplotr/

6.1 Annex 1: Quantitative results

6.1.1 SEROLOGY ON SERUM

Sample PT2022PRRSSERSERUMNS1

Lab number	97505-1	97505-2	97507	97508	97509-1	97509-2	97540
Method	M ₁	M ₂	M ₁	M ₁	M ₃	M ₁	M ₁
OD1	0.053	0.153	0.091	0.059	0.154	0.063	0.048
OD2	0.054	0.149	0.087	0.061	0.156	0.063	0.045
Mean	0.054	0.151	0.089	0.060	0.155	0.063	0.047
SD	0.001	0.003	0.003	0.001	0.001	0	0.002
CV (%)	1.85	1.99	3.37	1.67	0.65	0	4.26

(OD = optical density; SD = standard deviation; CV = coefficient of variation)

(M₁ = IDEXX PRRS X3 Ab; M₂ = Indical [Qiagen] – pigtype PRRSV Ab; M₃ = BIOCHECK – PRRS XR)

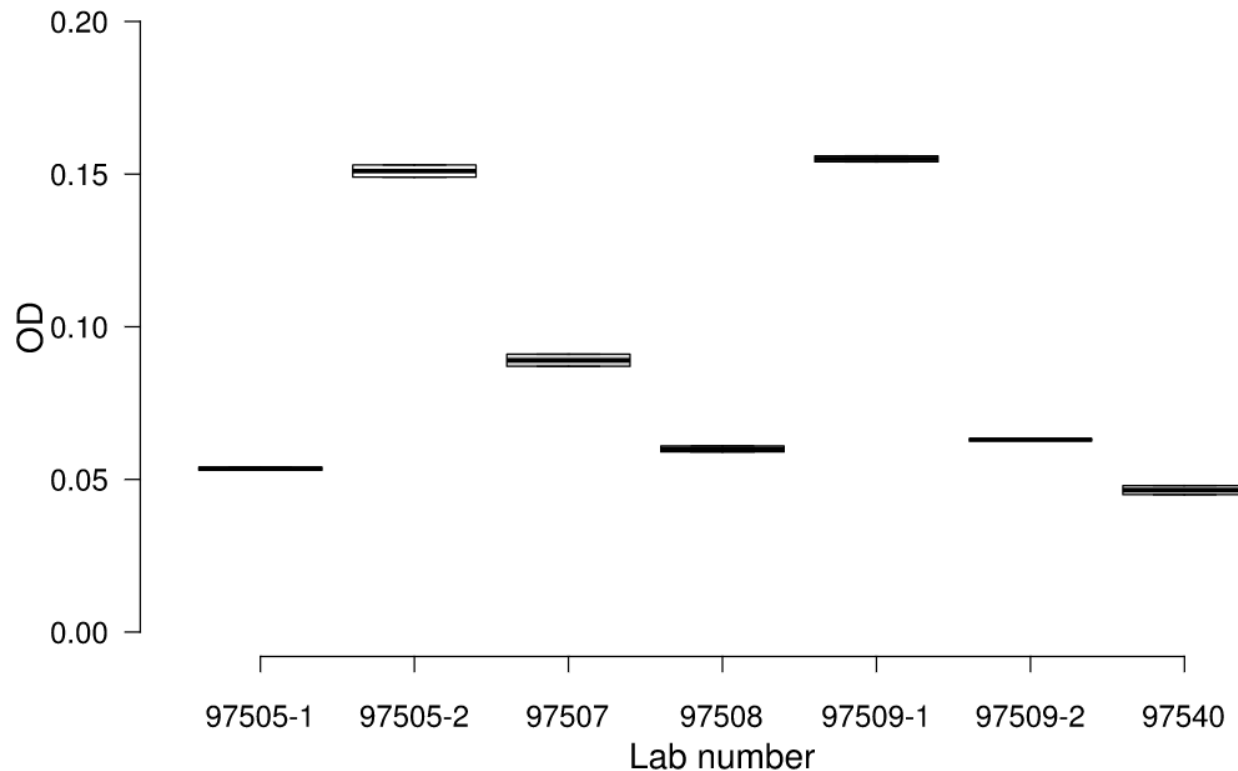


Figure 1. Distribution of the optical densities (box-plots) per laboratory

Lab number	97505-1	97505-2	97507	97508	97501	97509-2	97540
Method	M ₁	M ₂	M ₁	M ₁	M ₃	M ₁	M ₁
OD1	0.057	0.143	0.095	0.068	0.178	0.061	0.051
OD2	0.055	0.138	0.089	0.066	0.180	0.060	0.045
Mean	0.056	0.141	0.092	0.067	0.179	0.061	0.048
SD	0.001	0.003	0.004	0.001	0.001	0.001	0.004
CV (%)	1.85	2.13	4.35	1.49	0.56	1.64	8.33

(OD = optical density; SD = standard deviation; CV = coefficient of variation)
 (M₁ = IDEXX PRRS X3 Ab; M₂ = Indical (Qiagen) – pigtype PRRSV Ab; M₃ = BIOCHECK – PRRS XR)

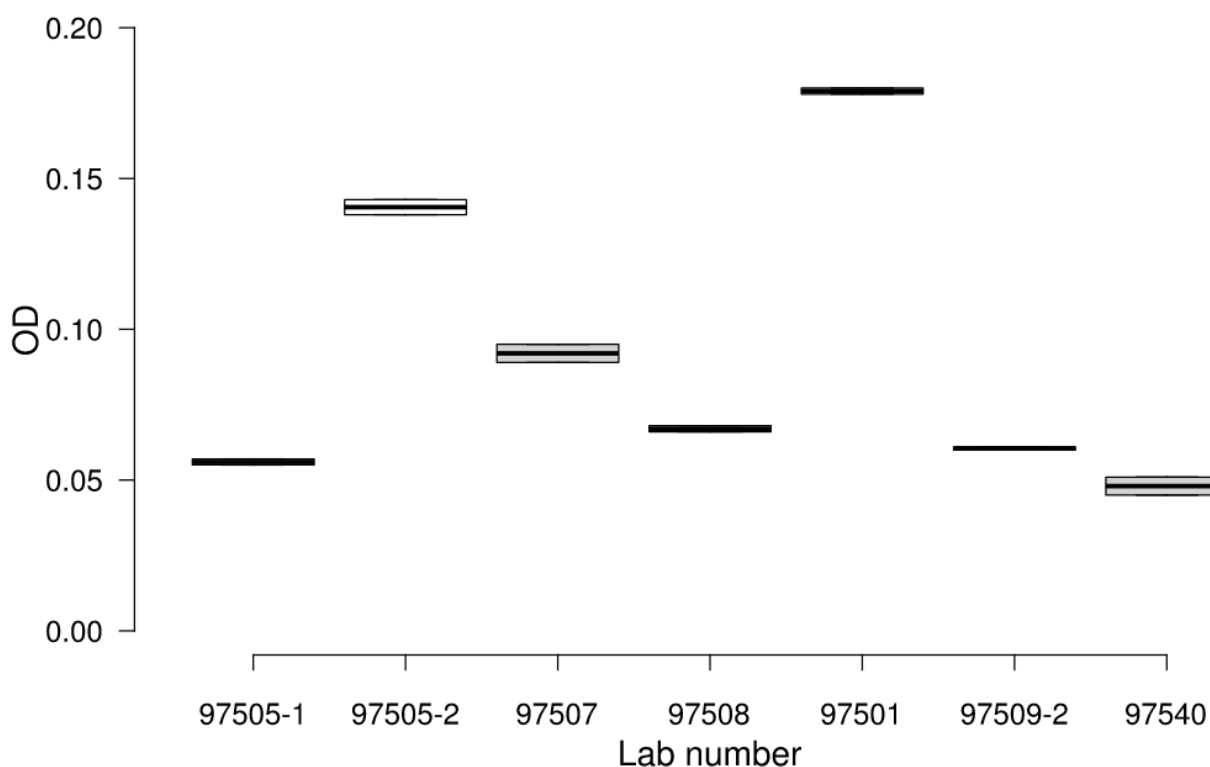


Figure 2. Distribution of the optical densities (box-plots) per laboratory

6.2 Annex 2: Additional information

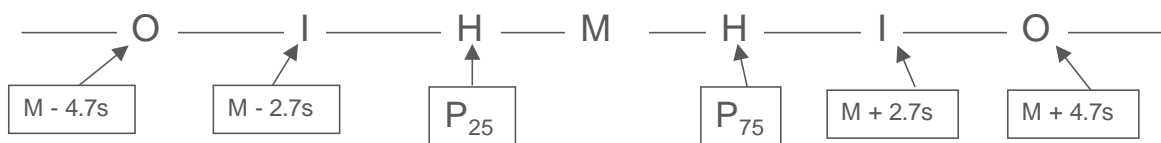
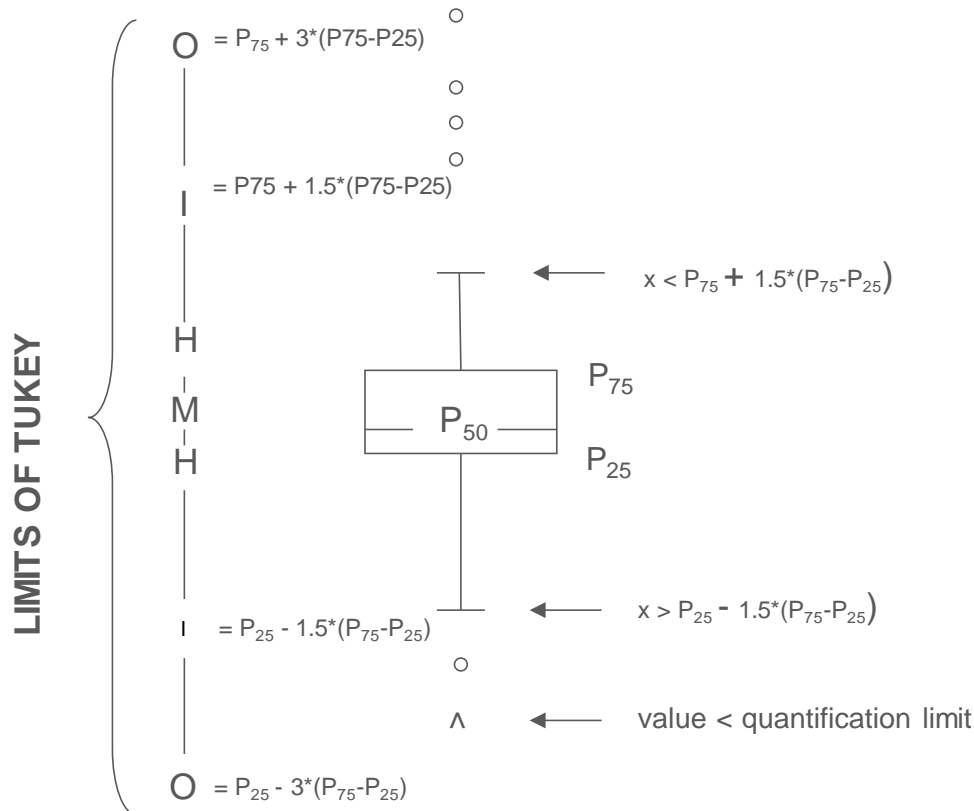
The preliminary report of this survey is available on our website via the following link:
https://www.wiv-isp.be/QML/activities/PT%20VET/fr/originaux/rapports_annee.htm

The calendar for Proficiency Testing in Veterinary diagnosis is available on our website:
https://www.wiv-isp.be/QML/activities/external_quality/calendar/kalender.htm

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

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