BIOLOGICAL HEALTH RISKS
QUALITY OF LABORATORIES

CLINICAL BIOLOGY
COMMITEE OF EXPERTS

EXTERNAL QUALITY ASSESSMENT
IN CLINICAL BIOLOGY

DEFINITIVE GLOBAL REPORT

Molecular Microbiology

Bordetella pertussis

SURVEY 2022/9

Sciensano/Molecular Microbiology/Bordetella pertussis/2-E

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

www.sciensano.be

ISSN: 2294-3498

COMMITTEE OF EXPERTS

Sciensano					
Secretariat		PHONE:	02/642.55.21 (Nl) 02/642.55.21 (Fr)	FAX:	02/642.56.45
		e-mail:	QL.secretariat@sciensano.be		
Bernard China	Scheme coordinator	PHONE:	02/6425385		
		e-mail:	Bernard.china@sciensano.be		
Kris Vernelen	Alternate coordinator	PHONE:			
		e-mail:	Kris.vernelen@sciensano.be		
Experts	**Institute**				
Marijke reynders	**AZ Sint Jan, Brugge**				
Samy	**CHU de Liège**				
Stefanie Desmet	**KUL**				
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Walter Verstrepen	**ZNA**				

A draft version of this report was submitted to the experts: 30/01/2023

Authorization of the report: by Bernard China, scheme coordinator

Signature of the scheme coordinator.

Date of publication: 23/02/2023

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INTRODUCTION

Sciensano organized in collaboration with the national reference laboratory (UZ Brussel) an External Quality Assessment (EQA) for the detection of *Bordetella pertussis* in sputum using molecular methods.

1.1 The samples

A strain of *Bordetella pertussis* was provided by the NRC and growth in Sciensano on charcoal agar. A suspension was made and titrated. A suspension at 5 10^8 CFUs/mL was made.as a master solution. From this suspension dilutions were made in sputum. 3 samples were prepared: a negative sample (sputum without bacteria) called BP22-1; a sample made of 500 µl of mother solution in 50 mL of sputum and then aliquoted to produce 30 samples of 1.5 mL. This sample was called BP22-2. 500 ul of BP22-2 was added to 50 mL of sputum and aliquoted (30x1,5 mL) to make sample BP22-3.

1.1.1 HOMOGENEITY

The samples were tested by the reference laboratory before sending to the participants to determine the target value. The samples were considered as homogeneous..

1.1.2 STABILITY

Stability was verified by comparing the results obtained by the reference laboratory before and during the investigation. Qualitatively the results are identical. We considered the samples to be stable.

1.2 Evaluation

For Qualitative detection, participants' results will be compared to the pre-survey results of the reference laboratory. The QCMD scoring system will be applied (Table 2).

Sample status	Score in case of correct answer	Score in case of wrong answer
Negative	0	+3
Frequently detected*	0	+3
Detected*	0	+2
Infrequently detected*	0	+1

* : Frequently detected : sample detected by more than 95% of the participants, detected : sample detected by more than 65% of the participants, infrequently detected : Sample detected by less than 65% of the participants.

1.3 Survey Timeline

Samples shipment: 6/12/2022
Deadline for the encoding of the results: 23/12/2022
Preliminary report: 02/01/2023 (on line: EQA Mol bio Micro - Preliminary report - 2022-9 | sciensano.be)

2 RESULTS

2.1 The participants

23 laboratories were registered and 21 (91.3%) return results.

2.2 Results per sample

Table R1. Results per sample

Sample ID	Status	Positive	Negative
BP21-1	Negative	0	21
BP22-2	Frequently detected	21	0
BP22-3	Frequently detected	21	0

100% of the encoded results were correct

2.3 Results per method and per laboratory

Table R2. Results per sample and per method for each laboratory

Method	target	BP2201	BP2202	BP2203	Score
Aries Bordetella kit	ptxA gene	0	1	1	0
Aries Bordetella kit	ptxA gene	0	1	1	0
Biofire film assay	ptxP	0	1	1	0
Biofire respi 2.1 plus panel	ptxP	0	1	1	0
Bordetella Elite MGB kit	ptxA gene	0	1	1	0
Bordetella Elite MGB kit	IS481; ptxA gene	0	1	1	0
Diasorin Simplexa Bordetella pertussis	IS481; IS1001	0	1	1	0
Qiastat-Dx respiratory panel	IS481	0	1	1	0
RTqPCR in house	IS481	0	1	1	0
RTqPCR in house	IS481	0	1	1	0
RTqPCR in house	IS481; IS1002	0	1	1	0
RTqPCR in house	IS481; ptxA gene	0	1	1	0
RTqPCR in house	IS481	0	1	1	0
RTqPCR in house	ptxS1; IS481	0	1	1	0
RTqPCR in house	IS481	0	1	1	0
RTqPCR in house	IS481 + 16S	0	1	1	0
Seegene Allplex	IS481	0	1	1	0
Seegene Allplex Pneumobacter Assay	IS481	0	1	1	0
Seegene Allplex Pneumobacter Assay	IS481	0	1	1	0
Seegene Allplex RP4	IS481	0	1	1	0
Seegene Allplex RP4	IS481	0	1	1	0

O: negative result; 1: positive result

2.4 Conclusions

All the participants encoded correct results. It is maybe due to the fact that the proposed positive samples were heavily contaminated. Eight of the 21 participants (38.1%) used a homemade PCR method. 5 participants (23.8%) used a multiplex method from Seegene. The target sequence was mostly IS481(16/21=76.2%) and the promoter region of the pertussis toxin gen. PCR on IS1001 was used to discriminate between *B. pertussis* and *B. parapertussis*.

END

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