EUROPEAN COMMISSION

View in the context of the Performance Evaluation Consultation Procedure (PECP)

Expert panels on medical devices and in vitro diagnostic devices (Expamed)

Contents

Scope of this expert view

This scientific view reflects the opinion of independent experts (MDR Article 106.1) on the performance evaluation report (PER) of the manufacturer. The advice is provided in the context of the performance evaluation consultation procedure (PECP), which is an additional element of conformity assessment by notified bodies for specific high-risk *in vitro* diagnostic devices (IVDR Article 48.6).

When making its conformity assessment decision, the notified body is obliged to give due consideration to the opinions expressed in the scientific view of the expert panel, where applicable (Annex IX, Section 4.9 or, as applicable, Annex X, Section 3, point (j)).

For class D devices, the notified body must provide a full justification in the case of divergent views between the notified body and the experts. This justification shall be included in the notification to the competent authority (IVDR Article 50; mechanism for scrutiny of class D devices).

1 ADMINISTRATIVE INFORMATION

Date of reception of the dossier	4/11/2021
Notified Body Number	2797
Internal PECP dossier #	IVD-2021-000010
***In vitro* diagnostic medical device**	This test is an in vitro nucleic acid amplification test intended for qualitative detection of SARS-CoV-2 genomic RNA by real-time polymerase chain reaction (PCR) method.

2 INFORMATION PROVIDED BY THE NOTIFIED BODY

When consulting the IVD expert panel, the notified body provided the below information on the type of device in accordance with MDCG 2021-22.

Intended purpose (P)		
P1	what is detected and/or measured *please specify the analyte(s) or marker(s), e.g. SARS-CoV-2 spike protein, Kel1 (K)*	SARS-CoV-2 RdRp, E and N genes
P2	function of the device *e.g. diagnosis, aid to diagnosis, monitoring, determining the infectious load, tissue typing etc*	Diagnostics and screening of SARS-CoV-2 and aid to diagnosis of COVID-19
P3	the specific disorder, condition or risk factor of interest that it is intended to detect, define or differentiate *e.g. hepatitis C infection, exposure to SARS-CoV-2, risk of HIV transmission in blood transfusion etc.*	COVID -19 disease, exposure to SARS-CoV-2
P4	whether it is automated or not	Manual
P5	whether it is qualitative, semi-quantitative or quantitative	Qualitative
P6	type of specimen(s) *e.g. whole blood, serum, saliva etc*	RNA extracted from nasopharyngeal swab (in transport media: UTM (Copan), PBS or Physiological saline solution); anterior nasal swab

P7	where applicable, the testing population *e.g. persons with specific health conditions, persons with specific symptoms, children in a certain age range*	EU population; Indicated and preventive testing of COVID-19
P8	intended user	Trained staff in laboratories
Technology (T)		
T1	principle of the assay method or principles of operation of the instrument *e.g. real-time PCR, qualitative PCR, digital PCR, sandwich immunoassay, competitive immunoassay, immunoturbidimetric assay etc.*	Real-time polymerase chain reaction (PCR) – amplification of the specific Target Sequence and detection using probes with fluorophore-based detection

3 VIEWS OF THE EXPERT PANEL

3.1 Information on panel and sub-group

Date of views	20/01/2022
Expert panel name	IVD expert panel
Sub-group of expert panel	IVD sub-group 2021-10

3.2 Summary of expert panel views

The assay is a reagent kit that is intended for the qualitative screening, diagnosis, and aid to diagnosis of SARS-CoV-2 infection and COVID-19. The kit is composed of a ready-to-use master mix, a positive control, and an internal control for the whole diagnostic process and can be run in diverse validated amplification instruments, with or without a previous RNA extraction step. The technology is based on SARS-CoV-2 RNA amplification by real-time PCR targeting *RdRp*, *E* and *N* genes in nasopharyngeal swabs and anterior nasal swabs that are preserved in transport media. The intended users are laboratory professionals specifically trained in the techniques of real-time PCR.

The scientific validity report was sufficiently comprehensive to support the association between SARS-CoV-2 genomic detection by real-time PCR and SARS-CoV-2 infection in different specimens. Evidence and recommendations were collected through a scientific literature review based on an adequate literature search methodology and were retrieved from accredited data bases of scientific publications and web pages of relevant international and national public health institutions (WHO, CDC).

The analytical report was sufficient to prove that the assay can detect the targeted analyte. In general, the number of samples or replicates that were used and the data analyses that were performed were adequate, criteria for acceptance of values of analytical parameters were defined and reported values of analytical parameters were acceptable. Of note, some shortcomings were detected in the assessment of precision, interference and stability performance that could result in overestimated values of assay precision.

The clinical performance report demonstrated the ability of the assay to yield results that were correlated with SARS-CoV-2 infection in common population screening. Reported values of clinical sensitivity and clinical specificity were optimal and statistically robust. However, results of assay clinical performance only obtained using nasopharyngeal swabs and implementing a previous automated viral RNA extraction step. No evidence were provided about assay clinical performance using anterior nasal swabs or implementing manual viral RNA extraction or direct viral RNA amplification methods.
The approach taken by the manufacturer to gather clinical evidence adequately addressed demonstration of scientific validity, analytical performance, and clinical performance. An appropriate literature search methodology was implemented to assess scientific validity and the search was made into reliable sources of information (accredited data bases of scientific publications and web sites of relevant public health institutions). The real-time PCR technology on which the assay is based is well known and has extensively been implemented for detection of SARS-CoV-2 since the virus emerged, as well as with other pathogens in the last decades. Clinical evidence provided was acceptable and was supported by robust results of analytical and clinical performance studies.

Evaluation of performance was nearly complete. The number of samples or replicates that were used to demonstrate analytical and clinical performance was sufficient, the data analyses that were performed were adequate, the criteria for acceptance of values of analytical parameters were defined and reported values of analytical and clinical parameters were acceptable, despite some shortcomings. The PMPF only reported one safety event, which provides consistent evidence of safe assay performance.

In general, the information of the PER provided sufficient clinical evidence of scientific validity, analytical performance, clinical performance, and safety of the assay. It is recommended that the manufacturer be requested to assess assay precision at LoD borderline concentrations, with a wider number of replicates, and over several days. In addition, the manufacturer should provide evidence on clinical performance using anterior nasal swabs and implementing manual viral RNA extraction or direct viral RNA amplification.

3.3 Views on the specific reports included in the performance evaluation report (PER)

(IVDR, Annex XIII, Section 1.3.2, first paragraph)

Views of the expert panel on the performance evaluation report of the manufacturer (PER)
1. Expert views on the scientific validity report[1]
The scientific validity report was comprehensive to support the association of SARS-CoV-2 genomic detection by real-time PCR with SARS-CoV-2 infection. Evidence and recommendations were collected through a scientific literature review based on an adequate selection of search terms and were retrieved from accredited data bases of scientific publications and web pages of relevant international and national public health institutions (WHO, CDC). The epidemiology and pathogenesis of SARS-CoV-2 infection were sufficiently described, as well as the association of the analyte with clinical conditions,

[1] Annex XIII, Section 1.2.1 of Regulation (EU) 2017/746- Demonstration of the scientific validity

and the adequacy of real-time PCR methods to detect SARS-CoV-2 in a wide diversity of specimens and, particularly, in upper respiratory tract samples. The selection of *RdRp*, *E* and *N* genes as targets for SARS-CoV-2 was appropriate and supported by WHO and CDC recommendations. However, the number of scientific publications cited across the report was limited (n = 15).

2. Expert views on the analytical performance report[2]

The analytical report assessed the following analytical parameters:

i. Limit of detection (LoD). Testing of 5 dilution series of SARS-CoV-2 RNA NIBSSC 20/146 concentrations from 500 to 20 IU/mL on 24 replicates yielded LOD values of 216.78 IU/mL (95% CI 149.37 – 430.63 IU/mL) using manual RNA extraction, 279.28 IU/mL (95% CI 206.30 – 474.70 IU/mL) using automated RNA extraction, and 417.97 IU/mL (95% CI 313.87 – 661.17 IU/mL) by direct virus RNA amplification). LoD values were calculated with 95% probability by probit analysis.
ii. Analytical specificity. A wide panel of 38 targets was assessed in 3 replicates, including SARS-CoV-2 and its variants alpha and delta, as well as and other pathogens with similar genetic structure or causing similar infections. No cross reactions were observed, except for SARS-CoV-1, in panel testing and in in-silico analyses.
iii. Analytical interference. Diverse potential interfering substances at levels set according to CLSI and FDA literature were added to negative transport medium spiked with SARS-CoV-2 positive control at 3x LoD. Of all tested substances, only budesonide (100 µg per tested sample) showed partial interference using a direct RNA isolation kit.
iv. Cross-contamination. 5 runs of positive and negative samples were performed without signs of cross-contamination.
v. Precision. Positive controls were diluted into concentrations at 7xLoD and 3xLoD and tested in 3 replicates. CV values for repeatability and reproducibility were ≤ 2.45.
vi. Stability. The kit components are stable for 12 months under the compliance with the manufacturer specified storage conditions (-20ºC ± 5ºC) and after 3 thawing cycles performed over the last 30 days.
vii. Total system failure. A 100% success for detection of 3x LoD spiked samples was reported for the assay with manual RNA extraction method while 99% success was reported with automated RNA extraction and direct RNA amplification.
viii. Specimen and extraction validation. Tests of extraction procedures and materials were performed on positive and negative nasopharyngeal swab samples in different transport media (PBS, PSS, UTM) as well as nasal swabs to validate the functionality of the reagent kit combined with different RNA extraction products. Furthermore, stability of the RNA extract at -20°C and at +5°C was analysed.

In general, the number of samples or replicates that were used to demonstrate analytical performance was sufficient, the data analyses that were performed were adequate, the criteria for acceptance of values of analytical parameters were defined and reported values of analytical parameters were acceptable. Of note, some shortcomings were detected in the assessment of precision performance.

[2] Annex XIII, Section 1.2.2 of Regulation (EU) 2017/746- Demonstration of the analytical performance

First, assessment was performed considering concentrations at 7xLoD and 3xLoD while CLSI literature recommends using LoD borderline concentrations.
Second, the number of 3 replicates tested at these concentrations was rather small (FDA recommends 5 determinations at each concentration).
Third, reproducibility was only assessed considering inter-lot variability and inter-operator variability, but between-day variability after testing over several days, which best ensures a realistic estimate of precision, was not specified.
Fourth, storage stability of extracted RNA over 24h was analysed for +5°C and -20°C together. When analysed separately for the temperature conditions significant differences up to 1 Ct value could be observed especially in magnetic separation or direct isolation after 4h of storage.
Fifth, interference testing using the vendor universal PCR/ control gave partial interference for neomycin/bacitracin, but not for Budesomide, as mentioned.
Altogether, these shortcomings could result in overestimated values of assay precision.

3. Expert views on the clinical performance report[3]

The clinical performance report demonstrated the ability of the assay to yield results that were correlated with asymptomatic and symptomatic SARS-CoV-2 infection in common population screening. Evidences were gathered on the basis on diverse sources of information: i) three single-centre clinical performance studies comparing the assay against similar CE IVD RT-PCR kits for the intended use of SARS-CoV-2 screening, using anonymous/left-over nasopharyngeal samples preserved in transport media and running a previous automated RNA extraction step (162 positive and 557 negative samples in total); ii) scientific peer-reviewed literature describing clinical performance studies of similar assays; and iii) published experience gained through routine diagnostic testing using results of external quality control panels from two different organizations (28 positive and 13 negative samples in total). The reported clinical sensitivity and clinical specificity values were 100.00% (95% confidence interval 97.11-100.00%) and 100.00% (95% confidence interval 99.15-100.00%), respectively, against similar CE IVD RT-PCR comparator assays. The manufacturer has not considered in the calculations the invalid results obtained with the technology, or a discrepant result (false positive, that was confirmed by another PCR test). These results in a better value of sensitivity and specificity. The same optimal values of sensitivity and specificity were obtained in the external quality control panels.

The number of positive and negative clinical samples that were used to demonstrate clinical performance for screening was sufficient, the data analyses that were performed were adequate, the criteria for acceptance of clinical sensitivity and clinical specificity values were defined, clinical performance of the assay was evaluated against similar CE IVD state-of-the art real-time PCR reagent kits targeting SARS-CoV-2, and values reported for these parameters were optimal. However, no evidence were provided about the assay clinical performance using anterior nasal swabs or implementing manual viral RNA extraction or direct viral RNA amplification methods. Also, data for the asymmetric LOD of the multiplex 3 target PCR or a comment why it could not be provided, were not documented.

Within the provided performance evaluation plan pages 5-9 are missing. According to the table of contents, they include the chapters “intended use”, “device description”, “state of the art”. The

[3] Annex XIII, Section 1.2.3 of Regulation (EU) 2017/746- Demonstration of the clinical performance

manufacturer user instruction of the kit is missing too. These “instructions of use” would provide a brief overview over the kit requirements and the performance claimed.

3.4 Views on specific assessment aspects of the performance evaluation report (PER)

(IVDR, Annex XIII, Section 1.3.2, second paragraph)

Views of the expert panel on the specific aspects included in the performance evaluation report of the manufacturer (PER)
1. The justification for the approach taken to gather the clinical evidence
The approach taken by the manufacturer to gather clinical evidence addressed sufficiently the demonstration of scientific validity, analytical performance, and clinical performance. An appropriate literature search methodology was implemented to assess scientific validity and the search was made into reliable sources of information (accredited data bases of scientific publications and web sites of relevant public health institutions). Evidence of analytical performance were reported for all parameters applicable for a qualitative assay. Evidence of clinical performance were gathered from three single-centre clinical performance studies, scientific peer-reviewed literature describing clinical performance studies of similar assays, and results of external quality control panels.
2. The literature search methodology, protocol and report
The scientific validity report implemented an adequate literature search methodology to gather evidence and the search was made into reliable sources of information (accredited data bases of scientific publications and web sites of relevant public health institutions). However, the number of scientific publications cited across the report was relatively small (n = 15).
3. The technology on which the device is based, the intended purpose of the device and any claims made about the device's performance or safety
The real-time PCR technology on which the assay is based is well known and has extensively been implemented for detection of SARS-CoV-2 since the pandemic onset, as well as to detect other pathogens in the last decades. The PMS monitoring activities were well described and only one safety event was reported since assay commercialization, which ensures safe assay performance. The possibility to use the assay directly on respiratory upper respiratory tract samples, circumventing viral RNA extraction, is innovative but was not demonstrated in the clinical performance studies.
4. Acceptability of clinical evidence (clinical data and performance evaluation results) against state of the art in medicine
Clinical evidence provided was acceptable and was supported by sufficient results of scientific validity, analytical performance, and clinical performance. The number of samples or replicates that were used to demonstrate analytical and clinical performance was sufficient, the data analyses that were performed were adequate, the criteria for acceptance of values of analytical parameters were defined and reported values of analytical and clinical parameters were acceptable for the intended qualitative uses, despite some shortcomings.

5. Adequacy of PMPF report(s), where applicable
The manufacturer reported only one safety corrective action that was applied to the kit in June 2021 (one tube of the internal control was found to have an incorrect primary label - blank label without printing). No other safety issues or vigilance events were reported for the assay from the date of market introduction up to the present. The monitoring activities included in the PMS are appropriate and ensure safe performance of the assay in operational conditions.

3.5 Overall conclusions and recommendations

Overall conclusions and recommendations on the performance evaluation report
In general, the information of the PER provided sufficient clinical evidence of scientific validity, analytical performance, clinical performance, and safety of the assay. Information provided supports the clinical benefit of the intended uses. It is recommended that the manufacturer provide further information on the following aspects: i) Results of assay precision at LoD borderline concentrations, with a wider number of replicates, and over several days. ii) Results of assay clinical performance using anterior nasal swabs for the intended uses of screening, diagnostics, and aid to diagnostics. iii) Results of assay clinical performance when implementing manual viral RNA extraction and direct viral RNA amplification, for the intended uses of screening, diagnostics, and aid to diagnostics. iv) Actualization of the information to the stability of extracted RNA and of the interference depended on the protocol used. v) Include the missing part of the performance evaluation plan (pages 5-9) and the package insert/ instruction for use into the PER.

3.6 Stakeholder information, where available

Relevant information provided by stakeholders, if applicable[4]
Has the Secretariat provided information from stakeholders?
☐YES ☒NO
If yes, please summarise the information and how itwastaken into account.
Not relevant

[4] According to Article 106.4 of Regulation (EU) 2017/745, expert panels shall take into account relevant information provided by stakeholders including patients' organisations and healthcare professionals when preparing their scientific opinions.

3.7 Divergent positions in case no consensus can be reached

In case no consensus on the views can be achieved[5], please summarise divergent positions
Please indicate how many of the experts of the panel had divergent views
There were no divergent views

[5] According to Article 106.12 of Regulation (EU) 2017/745,when adopting its scientific opinion, the members of the expert panels shall use their best endeavour to reach a consensus. If consensus cannot be reached, the expert panels shall decide by a majority of their members, and the scientific opinion shall mention the divergent positions and the grounds on which they are based.