LETTER TO THE EDITOR

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Evaluation of the QIAstat-Dx Respiratory SARS-CoV-2 Panel for early diagnosis of COVID-19

Dear Editor,

An effective response to the SARS-CoV-2 that has caused the coronavirus disease 2019 (COVID-19) pandemic[1] requires rapid and accurate diagnostic testing. We evaluate the QIAstat-Dx® Respiratory SARS-CoV-2 Panel—a multiplex real-time polymerase chain reaction (RT-PCR) assay—against an E-gene RT-PCR assay[2] that successfully identified cases at the start of the COVID-19 pandemic in Singapore.[3] The QIAstat-Dx is a cartridge-based assay that integrates sample ribonucleic acid (RNA) extraction, reverse transcription, and a multiplex RT-PCR assay against 22 different respiratory pathogens (including SARS-CoV-2) in 1 self-contained system.[4-7] For the SARS-CoV-2 analyte, the assay targets both the E-gene and open reading frame 1a (ORF1a) regions of the viral genome. Prior work comparing different sets of primers and probes found that this E-gene primer combination was one of the most sensitive.[8]

This study received approval for the use of residual diagnostic samples, as well as a waiver of requirements for patient informed consent, from the Singapore General Hospital Institutional Review Board (#2020/2138).

We first compared the analytical limits of detection (LoD) of the QIAstat-Dx against the E-gene RT-PCR assay. A quantitated pool of SARS-CoV-2 positive oropharyngeal swab samples was serially diluted and run on both platforms. Probit regression analysis was used to calculate the theoretical limits of detection for each assay. The E-gene RT-PCR LoD was 223 E-gene copies/reaction and the QIAstat-Dx LoD was 193 E-gene copies/reaction, with no statistically significant difference at the 95% confidence level.

Next, we tested the analytical sensitivity of the QIAstat-Dx by assessing its ability to detect SARS-CoV-2 infection on archived oropharyngeal swab samples. The first positive samples of 40 confirmed COVID-19 patients detected on our E-gene RT-PCR assay were run on the QIAstat-Dx. These samples best represent swabs obtained when acutely infected patients present for medical care. The QIAstat-Dx reported 38 out of the 40 samples as positive, giving a diagnostic sensitivity of at least 95%. The 2 missed samples had high cycle threshold (Ct) values of 31.7 and 36.3, respectively, and the viral RNA may have inadvertently degraded with the freeze-thaw cycle inherent in using archived samples. Among the 40 samples, the Ct values ranged 13.4–30.0.

Lastly, we assessed the QIAstat-Dx for diagnostic specificity and cross-reactivity against other common respiratory tract pathogens. Twenty samples negative for SARS-CoV-2 were run on the QIAstat-Dx platform (Table 1). The QIAstat-Dx did not cross-react with the common respiratory tract pathogens tested, and did not react with known negative samples, giving a diagnostic specificity of 100%.

Our study was limited by the global shortage of testing reagents and the need to prioritise clinical testing. Within these limitations, the QIAstat-Dx demonstrated good overall performance for the SARS-CoV-2 analyte, with a diagnostic sensitivity of 95% (38/40 positive samples) and diagnostic specificity of 100% (20/20 negative samples). There was no cross-reactivity with the other common respiratory pathogens tested. In vitro studies by the manufacturer claim no cross-reactivity with SARS-CoV, available in the QIAstat-Dx Respiratory SARS-CoV-2 Panel Instructions for Use (Handbook, March 2020). The analytical limit of detection was similar to our existing E-gene RT-PCR assay. Our findings are consistent with a separate QIAstat-Dx study using prospectively collected nasopharyngeal swabs.[4]

The self-contained cartridge format of the QIAstat-Dx offers several advantages over conventional RT-PCR assays. It is easy to use, with no separate viral RNA extraction, reverse transcription and RT-PCR steps. This requires less training compared with conventional RT-PCR assays. Furthermore, having fewer handling steps reduces the risk of sample contamination. The run time of 70 minutes is similar to other rapid PCR assays, compared with an average of 3 hours for conventional RT-PCR assays. Also, the ability to test for other common respiratory pathogens that present similarly to SARS-CoV-2 allows the clinician to quickly identify the causative agent.

The major drawback of the QIAstat-Dx platform is the assay throughput. A 1-module instrument is only able to run 1 sample at a time; additional modules are required to run more samples simultaneously.

Overall, the QIAstat-Dx Respiratory SARS-CoV-2 Panel seems best suited to small numbers of urgent

Table 1. QIAstat-Dx Respiratory SARS-CoV-2 Panel does not cross-react with other common respiratory pathogens tested

Sample no.	Known result	QIAstat-Dx result
A	Influenza A	Influenza A (H3)
B	Human Metapneumovirus	Human Metapneumovirus A/B
C	Coronavirus 229E Influenza A (low positive)	Coronavirus 229E
D	Coronavirus OC43/HKU1	Coronavirus HKU1
E	Coronavirus NL63	Coronavirus NL63
F	*Mycoplasma pneumoniae*	*Mycoplasma pneumoniae*
G	Influenza B	Influenza B
H	Rhinovirus	Rhinovirus/Enterovirus
I	Adenovirus	Adenovirus
J	Parainfluenza Virus 1	Parainfluenza Virus 1
K to T	No pathogens detected	No pathogens detected

samples, when concurrent detection of other respiratory pathogens is desired. Rapidly identifying respiratory pathogens allows for COVID-19 patients to be isolated quickly, while also offering early diagnosis for other patients with respiratory infections.

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