PROTOCOL

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Diagnostic Performance of an Antigen Test for SARS-CoV-2 Infection (COVID-19)
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Introduction

Testing for SARS-Co-V-2-related disease is an important and complex topic during the current pandemic. There are a variety of testing solutions available, and the fact that most are released for clinical use under the FDA's Emergency Use Authorization means that the study of their accuracy needs to meet criteria that are less strict than usual. Of particular concern is testing of symptomatic patients, where accuracy is important and time-sensitive, and the need for more rapid-turnaround solutions is growing daily as commercial and public health labs face processing delays for the most-favored test, the real-time polymerase chain reaction-based assays (rtPCR). Antigen-based testing, increasingly available from commercial labs, promises a more rapid result – often within an hour of the test – but at a cost of lower sensitivity compared to PCR testing. The Sofia antigen test (Quidel, Inc.) is one of two antigen tests approved by the FDA with an Emergency Use Authorization. The FDA's EUA letter for the antigen test states that "Negative results should be treated as presumptive and confirmed with a molecular assay, if necessary, for patient management."

In addition, the ability to do these tests via mid-turbinate nasal swab (rather than by nasopharyngeal swabbing) would enable their use in a wider variety of settings with more limited personal protective equipment (i.e., limited availability of N95 respirators). There are only a few studies of the diagnostic performance of antigen testing available in the published and pre-print literature. The sensitivity of antigen testing in these studies varies considerably, and most of the studies were done using NP swabs (see Table and References). The purpose of this study is to compare the diagnostic performance of SARS-CoV2 antigen test obtained by mid-turbinate nasal swab with the reference standard rtPCR testing from nasopharyngeal swabs in a population of adult patients with symptoms consistent with COVID-19 disease.

Methods

Study design: Cross sectional diagnostic testing study

Subjects: Adults (>=18 years) with symptoms of COVID-19 infection (as determined by their treating physicians) for fewer than 5 days who are sent to a regional testing center for rtPCR testing by nasopharyngeal swab. Healthcare workers from a large regional health system are included in this population.

Setting: Outpatient

Testing Methods: The test of interest is a mid-turbinate nasal swab (both nares) using the Sofia rapid antigen test (Quidel, Inc.). The reference standard used will be a Quest rtPCR tests using nasopharyngeal swabbing technique of both nares. The antigen test will be collected first to reduce the chance that the detection will be inappropriately improved due to rhinorrhea produced by nasopharyngeal swabbing. If additional testing – such as influenza, RSV or streptococcal testing is ordered by the PCP, this will be performed after the antigen testing also.

Data Collection and Management: The antigen testing samples will be processed on-site at the regional testing centers, and the results and identifiers will be entered into a RedCap database for the study. The rtPCR test results will be processed at Quest Laboratories. Results of the rtPCR tests will be sent to the patient's ordering physician. The results for the enrolled patients will be extracted from the Epic EHR and matched to the antigen test results in the RedCap database.

Sample Size Determination: Because of operational concerns for healthcare workers, it is determined that the lowest acceptable sensitivity of the antigen test (relative to the rtPCR) is 90%. The average test positive rate in the testing centers for "Persons Under Investigation" for the month prior to the study was approximately 10%. A sample size of 800 subjects achieves 80% power to detect a difference of 0.099 between two diagnostic tests whose sensitivities are 0.999 and 0.900. This procedure uses a two-sided McNemar test with a significance level of 0.05000. The proportion of discordant pairs is 0.101.

Analysis: The results of the testing will be analyzed using SAS (Cary, NC). The following statistics will be calculated for the antigen test (relative to the reference standard rtPCR test): prevalence, sensitivity, specificity, positive and negative predictive values, likelihood ratios for positive and negative tests, and overall test accuracy.

Results

We anticipate presenting descriptive statistics and the diagnostic test characteristics as described in the Methods-Analysis section.

Discussion

We anticipate better understanding of the diagnostic value of SARS-CoV-2 antigen testing in the clinical management of the COVID-19 epidemic.

References

1. Lambert-Niclot S, Cuffel A, Le Pape S, Vauloup-Fellous C, Morand-Joubert L, Roque-Afonso A-M, et al. Evaluation of a Rapid Diagnostic Assay for Detection of SARS-CoV-2 Antigen in Nasopharyngeal Swabs. J Clin Microbiol [Internet]. 2020 Jul 23 [cited 2020 Aug 11];58(8). Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7383555/

2. Nagura-Ikeda M, Imai K, Tabata S, Miyoshi K, Murahara N, Mizuno T, et al. Clinical evaluation of self-collected saliva by RT-qPCR, direct RT-qPCR, RT-LAMP, and a rapid antigen test to diagnose COVID-19. J Clin Microbiol [Internet]. 2020 Jul 7 [cited 2020 Aug 11]; Available from: https://jcm.asm.org/content/early/2020/07/03/JCM.01438-20

3. Diao B, Wen K, Chen J, Liu Y, Yuan Z, Han C, et al. Diagnosis of Acute Respiratory Syndrome Coronavirus 2 Infection by Detection of Nucleocapsid Protein. medRxiv. 2020 Jan 1;2020.03.07.20032524.

4. Mak GC, Cheng PK, Lau SS, Wong KK, Lau C, Lam ET, et al. Evaluation of rapid antigen test for detection of SARS-CoV-2 virus. J Clin Virol. 2020 Aug;129:104500.

5. Scohy A, Anantharajah A, Bodéus M, Kabamba-Mukadi B, Verroken A, Rodriguez-Villalobos H. Low performance of rapid antigen detection test as frontline testing for COVID-19 diagnosis. J Clin Virol. 2020 Aug;129:104455.

6. Porte L, Legarraga P, Vollrath V, Aguilera X, Munita JM, Araos R, et al. Evaluation of novel antigen-based rapid detection test for the diagnosis of SARS-CoV-2 in respiratory samples. Int J Infect Dis [Internet]. 2020 Jun 1 [cited 2020 Aug 11]; Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7263236/

7. Blairon L, Wilmet A, Beukinga I, Tré-Hardy M. Implementation of rapid SARS-CoV-2 antigenic testing in a laboratory without access to molecular methods: Experiences of a general hospital. J Clin Virol. 2020 Aug;129:104472.

8. Mertens P, De Vos N, Martiny D, Jassoy C, Mirazimi A, Cuypers L, et al. Development and Potential Usefulness of the COVID-19 Ag Respi-Strip Diagnostic Assay in a Pandemic Context. Front Med [Internet]. 2020 May 8 [cited 2020 Aug 11];7. Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7227790/

9. Castro R, Luz PM, Wakimoto MD, Veloso VG, Grinsztejn B, Perazzo H. COVID-19: a meta-analysis of diagnostic test accuracy of commercial assays registered in Brazil. Braz J Infect Dis. 2020;24(2):180–7.

Table. Prior Diagnostic Testing Study results – review August 11, 2020

Study	Pre-print?	Ag test	Comp	Setting	Symptoms?	Results	Comments
Lamber-Niclot(1)		CORIS	rtPCR (several brands)	France, lab-based	Unclear (likely yes)	Sens – 50% Spec – 100% (sens 82.2% with Ct < 25)	
Nagura-Ikeda(2)		ESPLINE SARS-CoV-2	rtPCR (several)	Japan, inpatient	Diagnosed COVID-19 by swab – 15% asymptomatic. Range of duration of symptoms (up to 10 days).	Sens (antigen test) 11.2% in saliva specimen (rtPCR sensitivity from saliva test c/w NP swab rtPCR ranged from 50-81.6%)	self-collected saliva specimens tested with both PCR and Ag testing against NP swab PCR.
Diao(3)	Yes	Direct antigen test (NP swan)	rtPCR	Hospital	yes	Sens – 68%, spec – 100% (Ct <=40) sens 98%, spec 100% (Ct <=30)	Also tested urine antigen – not as good.
Mak(4)		BIOCREDIT COVID-19 (variety of sources – throat saliva, NP aspirate + throat swab, sputum)	rtPCR	Lab-based: samples from COVID positive patients	Yes	Sensitivities: Low+high viral load – 11.1-45.7% High viral load – 28.6-81.8% Low viral load – 0-21.1%	NP swab+throat saliva or NP aspirate+throat saliva had greater detection. Ct < 18.57 = high viral load.

Scohy(5)		CORIS	rtPCR	Belgium, hospital samples	30.4% asymptomatic, mean symptomatic time 4 days	Sens (overall) – 30.2% Ct < 25, sens 100% Ct < 30, sens 70.6% Ct <35, sens 46.9%	Mean Ct = 33
Porte(6)		Bioeasy Biotech Ag Test (NP+OP samples)	rtPCR	Respiratory emergency room at private hospital, Chile	Resp symptoms and/or fever with epi risk factor	Overall – sens 93.9%, spec 100% 0-7 d sxs – sens 94.7%, spec 100% 8-12 d sxs – sens 80.0%, spec 100% Ct <= 26 sens 100% Ct > 26 sens 72%	
Blairon(7)		CORIS Respi-Strip (from NP swabs)	rtPCR	General hospital	Not clear	Sensitivity 23.9%	Sent antigen samples from hospital to university center for PCR testing. No Ct-based analysis performed No symptoms or time from symptom onset reported.
Mertens(8) (clinical study)		CORIS respi-strip (from NPA, NPS, BAL samples)	rtPCR (several)	Multi-site retrospective sample study	yes	Overall Sens 57.6%, spec 99.5% Ct<25 sens 73.9%, spec 99.5% HCW overall sens 68%, spec 100% Ct<25 sens 92.9%, spec 100%	
Castro(9) Meta-analysis		Eco diagnostics	rtPCR	Unclear	Yes, unclear time since symptoms	Overall sens 89-91%, spec 87-93%	Specificities here lower than in any other study (?)

		(2 different tests)					Restricted to tests available in Brazil