Interpreting COVID-19 test results in clinical settings: it depends!

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ABSTRACT

Tests for COVID-19 are intended for a disparate and shifting range of purposes including: (1) diagnosing patients who present with symptoms to inform individual treatment decisions; (2) organizational uses such as “cohorting” potentially infected patients and staff to protect others; and (3) contact tracing, surveillance, and other public health purposes. Often lost when testing is encouraged is that testing does not by itself confer health benefits. Rather, testing is useful to the extent it forms a critical link to subsequent medical or public health interventions. Such interventions might be individual-level, like better diagnosis, treatment, isolation, or quarantine of contacts. They might aid surveillance to understand levels and trends of disease within a defined population that enables informed decisions to implement or relax social distancing measures. In this paper, we describe the range of available COVID-19 tests; their accuracy and timing considerations; and the specific clinical, organizational, and public health considerations that warrant different testing strategies. Three representative clinical scenarios illustrate the importance of appropriate test use and interpretation. The reason a patient seeks testing is often a strong indicator of the pre-test probability of infection, and thus how to interpret test results. In addition, the level of population spread of the virus and the timing of testing play critical roles in the positive or negative predictive value of the test. We conclude with practical recommendations regarding the need for testing in various contexts, appropriate tests and testing methods, and the interpretation of test results.

"Test, test, test" say public health experts, political leaders, and the media. After all, rapid and extensive testing was seen as the key to stopping the COVID-19 outbreak in South Korea and other countries in February, 2020.[1] Moreover, the lack of approved tests blinded health officials in Washington State and California to some of the first U.S. cases[2] and complicated the New York region's response to a surge of cases, hospitalizations, and deaths[3]. Testing capacity increased in May and June, but shortages re-emerged when cases surged in the South and West in July, causing long delays.[4]

In response to shortages, public health laboratories, hospitals, and private companies have developed a vast array of tests for diagnostic, public health, and other purposes. Each test, of course, has a different level of sensitivity and specificity, some not very high. These tests are intended for a disparate and shifting range of purposes including: (1) diagnosing patients who present with symptoms of COVID-19 to inform individual treatment decisions; (2) organizational uses such as "cohorting" potentially infected patients and staff to protect others; and (3) contact tracing, surveillance, and other public health purposes. The focus in this discussion is on the role of testing for individual patients and clinical decision-making, though we recognize there are several surveillance based purposes for testing that inform epidemiologic understanding of the virus.

Public health advisories, which early in the pandemic aimed at ensuring the few available tests were used where most needed, switched to encouraging more general testing of the population when tests became more readily available, but without clear guidance on appropriate use[5]. The public responded to increased availability of tests in complex and unpredictable ways, reflected in behaviors such as an increased demand for testing, social norms to get tested, or mistrust of testing and the purpose of test results. Often lost when testing is encouraged is that testing is not itself an intervention and does not by itself confer health benefits. Rather, testing is useful to the extent it forms a critical link to subsequent medical or public health interventions. Such interventions might be individual-level, like better diagnosis, treatment, isolation, or quarantine of contacts. Or they might focus on population health, such as surveillance to understand levels and trends of disease within a defined population that enables informed decisions to implement or relax social distancing measures.

As a result of this complex and ever evolving situation, it may be difficult for physicians to advise their patients on whether and how to be tested. Because of changing population factors, it is even challenging how to interpret the results of tests that have been performed. Is a patient concerned about symptoms she is experiencing or a possible recent exposure to someone who is infectious? Does she, or someone in her household, have underlying conditions that would put them at high risk if infected? Does his employer require "a COVID test" as a condition of returning to work? Has she been notified by the local health department that one of her co-workers has recently been infected?

To help physicians advise patients about COVID-19 testing and interpret the results, we first review the multiple purposes of testing at the individual, organizational, and population levels;

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the shifting demand for testing; and the characteristics of individuals tested in each of these settings. These factors are important because in settings where the prevalence of infection is low, a positive test may be more likely to be a false positive than indicative of a true infection. We then describe the range of available tests; their accuracy and timing considerations; and the specific clinical, organizational, and public health considerations that warrant different testing strategies. The final section of this paper uses three representative scenarios to illustrate the importance of appropriate test use and interpretation. We conclude with practical recommendations in various contexts regarding the need for testing, appropriate tests and testing methods, and the interpretation of test results.

The who, why, and how of COVID-19 testing

What are the purposes of testing for COVID-19?

Typically, tests are conducted for two primary purposes: diagnosis of persons with clinical symptoms and surveillance of disease prevalence in the population. Historically, there has been a greater emphasis on testing for diagnostic rather than surveillance purposes, especially in the absence of widespread outbreaks. COVID-19 is atypical in this regard. With a threat like COVID-19, the purposes of testing become more nuanced and vary for individuals, organizations, and government actors. Attention to these reasons can help physicians anticipate when demand for testing may increase due to individual, organizational, or population level pressures, and when increased counseling of patients may be necessary.

Who gets tested?

Individual motivations, clinical choice, organizational policy, and population determinants influence which individuals actually receive a test for COVID-19.

Individual purposes

At the individual level, the purpose of testing is tied to the perceptions of the clinician and the possible patient. What would the clinician do differently for the patient and themselves based on testing capacity or results? Clinicians may recommend different courses of action if they are able to diagnose a case of COVID-19 depending on the patient's underlying conditions. Monitoring of the individual and the individual's family may be different if the patient were to be diagnosed with COVID-19 rather than influenza or another similar condition, especially if patients or their family members fall into a high-risk group such as pregnant or immunocompromised[6]. Clinician behavior might also vary based on the ability to test and/or the test result[7]. For instance, they may need to modify their use of PPE and other personal protections in the event they come into contact with a presumed-positive patient.

From the patient perspective, psychological motivations and setting influence test seeking behavior. Motivations to seek testing are varied but are likely influenced by known constructs based on theoretical models of health behavior (Table 1). These constructs include perceived susceptibility for infection, perceived severity of disease, perceived benefits and barriers to getting tested, cues to action[8], and self-efficacy. For example, if an individual lives in a geographic area where others have had COVID-19 (perceived susceptibility), has seen others experience symptoms (perceived severity), wants to protect family members (perceived benefits) or negatives of the test (e.g. getting it is uncomfortable), started showing symptoms (cue to action) or works in profession where testing is being encouraged or as part of a public health strategy to protect the community as a whole (cue to action), and believes they have control over accessing a test and protective behaviors as a result of the test (self-efficacy), testing is a logical action.

External factors such as the need to travel or return to work expectations can also be cues to action. Some people, often referred to as the "worried well," self-refer to medical care out of a desire to affirm that clinical care is not needed or alleviate a level of dread associated with an event[9]. Local testing capacity and the affordability of tests can either facilitate or be a barrier to testing. Early in the pandemic, when testing resources were not available in sufficient supplies, many were denied the opportunity to be tested, even if they exhibited some symptoms or were likely to have been exposed. Later, when testing did become widely available, some may have sought it because its earlier unavailability made it seem desirable[10]. Particular demographic groups may be more likely to be tested because their cues to action are greater based on their presumed exposure or level of risk due to their occupation.

Test results could also influence subsequent behavior. Despite policy recommendations in many geographic areas to physically distance from others and stay home regardless of clinical confirmation of COVID-19, individuals may change their behavior after the confirmation of either a positive diagnosis for an active infection or a positive result on an antibody test. Determining antibody status, in particular, has been incentivized by political discussions of immunity passports, possibly creating perverse incentives for getting infection and antibodies[11].

Organizational level purposes

At the organizational level, routine testing for SARS-CoV-2 may be useful in settings where it is not possible to work remotely in order to quickly isolate infected workers. The rationale for such testing in health care settings, where close proximity and physical contact is often unavoidable, is to avoid having asymptomatic or pre-symptomatic patients and health care workers inadvertently transmit the virus to others. For similar reasons, other non-health care groups that would be high priority for universal testing include prisoners and prison employees; workers at warehouses, factories and food processing plants; and certain other types of public employees (police, firefighters, road maintenance, sanitation workers). Such testing may have prevented or

mitigated large outbreaks such as one at an Amazon warehouse in Pennsylvania where more than 100 workers have tested positive for COVID-19[12] or in meat packing plants and correctional facilities, which are important sources of infection.

Similarly, high-frequency testing is a cornerstone of many universities' strategies to be able to reopen at lower risk of large outbreaks. Modeling suggests that high-frequency testing with adequate contact tracing, isolation, and quarantine, may be able to suppress transmission on college campuses or similar settings[13], but that the tests most likely to have sufficiently fast turnaround time at low cost may also lead to many false positives. Interpreting test results in this context is highly dependent on the surveillance strategy employed and beyond normal clinical practice, so we do not go into detail about it.

Population-level purposes

At the population level, COVID-19 testing is intended to provide public health authorities and political leaders sufficient understanding of the epidemic to impose and relax control measures appropriately. Broadly speaking, testing for viral RNA or antigen is intended to provide information about the current incidence and prevalence and trends over time. Testing for antibody against SARS-CoV-2 is intended to help authorities understand whether there are sufficient levels of immunity that control measures can be relaxed[14]. To make these decisions, COVID-19 testing data should be coupled with additional population health information, such as evidence about hospitalizations and hospital capacity,[15] COVID-19-attributed mortality, excess mortality consistent with COVID-19,[16] and behavioral surveillance such as aggregated mobile phone movement data[17].

Factors that influence test interpretation are all constantly changing. For instance, more and different types of tests are being developed which have lower cost and shorter turnaround times, but may be less sensitive and specific. Testing resources which were in short supply early in the pandemic, became more available. Then, with the resurgence in southern and western states in June and July of 2020, shortages are emerging again[18]. With constrained resources, testing tends to be focused on individuals with symptoms or a likely exposure, and thus those with a higher likelihood of having COVID-19. For instance, New York City's criteria for COVID-19 testing were expanded on July 2, 2020 to include individuals who participated in demonstrations or other large gatherings within the preceding 14 days and those who plan to visit people who may be at risk of severe COVID-19 if infected. Similarly, there may be an increase in the number of people tested for surveillance purposes when businesses and schools re-open, possibly resulting in a lower test positivity rate (the proportion of all tests that find a positive result).

What types of tests exist?

COVID-19 testing has primarily focused on detecting the SARS-CoV-2 viral RNA, one of its structural proteins, or the antibodies the body produces in response to the virus.

Currently, there are two broad categories of tests available for SARS-CoV-2 infection: viral and serologic. Viral tests are designed to directly detect viral components (i.e. RNA or protein) during an active infection while serologic tests are designed to detect presence of antibodies. As of July 30, 2020, the FDA has approved 193 viral and serologic tests under the Emergency Use Authorization (EUA) which allows expedited approval without the usual level of review[19].

Test performance and test interpretation of the results, are described using the following standard measures:

- sensitivity (Se), the proportion of infected individuals who test positive
- specificity (Sp), the proportion of uninfected individuals who test negative
- positive predictive value (PPV), the probability that a positive result actually means one has COVID-19. Imperfect PPV means some percentage of isolation or treatment is unnecessary; for antibody tests it means some people will believe they are immune when they are not.
- negative predictive value (NPV), the probability that a negative result actually means one does not have COVID-19. Imperfect NPV means that some percentage of people told they do not need to isolate are in fact infectious.

Any test's positive and negative predictive value, however, also depends on the pretest probability of infection: the probability a person has COVID-19 in the absence of a test result[20]. Pretest infection probability is often thought of as the prevalence of disease in a person's geographic or demographic subpopulation, but it also varies within subpopulations based on exposures (a known COVID-19 contact has higher pretest probability of infection) and clinical information (as does a person with fever and dry cough). A person's motivation for seeking a COVID-19 test often results from their perceptions of their prior probability of infection and thus can be probative of predictive values. One point about test interpretation that is important to emphasize is that, even with reasonably high Se and Sp, but low prevalence or pre-test probability, PPV is likely to be low. This is simply because the number of false positives is large relative to true positive results[21, 22]. Thus, unless a patient has symptoms of COVID-19, or a reason to suspect recent exposure to an infectious person, a positive test result may not mean a true infection.

Viral Tests

Viral testing includes both rapid antigen and molecular tests. A rapid antigen test detects the presence of viral proteins to confirm the infection as quickly as 15 minutes (i.e. Quidel's Sofia). These tests are generally lateral flow immunoassays or, simply, strip tests that work in the same

way as a pregnancy test. Respiratory sample flows laterally down the strip and if target viral antigens are present, they bind to fluorescent antibodies that produce a visible fluorescent line on the strip, indicating a positive test result. Although rapid tests generally provide qualitative results, they can be linked to a device that analyzes the level of fluorescence to provide quantitative data as well (i.e. Quidel's Sofia Antigen FIA)[23]. In contrast, a molecular test, the more reliable of the two, relies on reverse transcription PCR to amplify and quantify the viral RNA present in the respiratory sample. Although both can diagnose an acute SARS-CoV2 infection, molecular testing is currently the preferred diagnostic test for SARS-CoV2 due to better testing performance, so this section will focus on molecular, not rapid antigen, tests. (Because antigen tests are substantially less expensive than PCR tests, they could likely play an important role in high-frequency testing to protect congregate settings like universities or prisons, but this is a special surveillance situation outside the scope of this paper [24, 25].)

Timing of testing relative to test accuracy

In severe and mild cases, viral RNA has been reliably detected as early as day 1 of symptom onset and throughout the first 7 days. While in severe cases the positive rates remained high 8-14 days after onset of symptoms, they have been found to be substantially lower in mild cases[26]. Therefore, testing for acute SARS-CoV2 infection should be done soon after symptoms arise, preferably within the first week after symptom onset, to ensure highest probability of accurately detecting the viral RNA (sensitivity). Molecular testing after resolution of symptoms is of questionable utility for most patients since a positive result may represent remaining RNA from nonviable viral particles and may not provide significant clinical value. As a result, CDC no longer recommends using negative PCR results to clear a patient from isolation or testing asymptomatic people within three months of a previous positive test result[27]. In contrast, an antibody test after resolution of symptoms may be reasonable to assess immunity--though there remains debate about to what extent the persistence of measurable antibody correlates with the extent and duration of immunity--or recommended for surveillance purposes to estimate cumulative incidence in a population[28].

Additionally, challenges with laboratory capacity often led to very long times between sample collection and patients and providers receiving results at points in 2020[29]. These delays were typically worst when a population was experiencing a large surge in COVID-19 cases--exactly the time when rapid, valid test results were most needed. Delays in testing reduce the effective sensitivity of a test. For purposes of suppressing transmission, sensitivity is effectively reduced proportional to the period of infectiousness during which the result is unknown, and for clinical purposes sensitivity is essentially zero if results are not known at the latest point a clinical decision would need to be made based on them. As a result, some jurisdictions recommend that patients quarantine until receiving test results on the assumption heightened risk led them to seek

a test and physicians often have to assume a patient has COVID-19 based on clinical criteria alone[30].

Test Performance

Test performance for molecular PCR tests depends on the type of specimen collected, time since infection, and the specific probes used to target a particular region of the viral RNA. Regarding specimens, sputum has been shown to have the highest sensitivity (up to 88.9%) during the first 14 days after illness onset followed by nasopharyngeal swabs (73.3%) and throat swabs (61.3%)[26]. However, since only a small fraction of infected individuals produce sputum, nasal swabs may be the most practical method of collecting specimens for testing. Regarding probes, private laboratories and CDC have developed probes targeting a variety of viral genes, commonly the envelope (E), nucleocapsid (N), and RNA-dependent RNA Polymerase (RdRp). Although sensitivities vary based on which probe and assay were used, specificity of molecular testing have been consistently high[31]. Furthermore, independent organizations evaluating commercially available tests show both sensitivity and specificity of majority of molecular tests are above 90%[32]. However, even with relatively high sensitivity and specificity, the predictive value of test results depends on the prevalence of disease. In a meta-analysis, the pooled PPV and NPV of molecular tests were estimated to be 47.3% and 99.9% at a disease prevalence of 1%, 90.8% and 98.8% at a prevalence of 10% and 98.3% and 93.4% at a prevalence of 39%, respectively[33]. (Based on studies extracted from China, Italy and Japan, the pooled prevalence was 38%. The pooled prevalence in just China was 39%). Additionally, a study found 21.4% of patients admitted for symptoms suspicious for COVID had two consecutive negative RT-PCR test results on different dates after symptom onset before eventually testing positive[34]. This suggests some may have longer nucleic acid conversion time and, thus, an initial negative test in those with clinical suspicion cannot completely rule out an infection.

No cross-reactivity with other respiratory viruses seem to have been observed with molecular testing for SARS-CoV2 as well[35, 31]. A study evaluating the specificity of RT-PCR probes tested 297 clinical samples from patients with a broad range of respiratory agents such as common human coronaviruses (HCoV-HKU1, OC43, NL63, 229E), influenza subtypes, rhinovirus, parainfluenza, etc. Testing yielded no false positive outcome and thus suggests there was no cross-reactivity[35].

Serologic Tests

Serologic testing targets the antibodies the body produces in response to the virus, specifically the viral spike glycoprotein and nucleocapsid phosphoprotein[36]. Currently, within serologic testing there are rapid antibody tests and automated immunoassays. Rapid antibody tests, similar to rapid antigen tests, can be a lateral flow assay (LFA) that uses fluorescent antibodies to

indicate whether target antibodies against SARS-CoV2 are present within the sample serum, plasma, or blood. In addition, there are automated immunoassays such as ELISA that can quantify the level of antibodies present for a more definitive analysis. Presence of ~~IgM and~~ IgG antibodies indicates the individual had a prior infection and thus likely has some level of immunity against the novel coronavirus. However, whether prior infection definitively protects against reinfection or how long such immunity persists is currently uncertain[37]. A recent Chinese study evaluating the immune response of COVID-19 patients found that more than 90% of both asymptomatic and symptomatic individuals had a reduction in virus-specific IgG antibodies in the early convalescent phase. In fact, 40% of asymptomatic individuals became seronegative while 12.9% of symptomatic individuals became negative for IgG in the convalescent phase[37]. This suggests that immunity from SARS-CoV-2 infection may be short-lived, especially in asymptomatic individuals. However, whether individuals are protected against re-infection while antibodies are present has yet to be studied. Similarly, immunity of SARS-CoV-1 and MERS-CoV is also unknown since SARS has not reemerged since 2004 and MERS cases remain sporadic. Reinfections are, however, seen with common human coronaviruses such as NL63 and OC43[38]. In other words, detection of IgG antibodies alone is not synonymous with durable immunity and should not guide public health measures until more data regarding long-term immunity against reinfection is available.

Timing of testing

Most infected individuals develop detectable levels of IgM and IgG by week 2 after symptom onset. A study evaluating seroconversion (the production of SARS-CoV2-specific antibodies) found that antibodies were detected in less than 40% of patients 1-week after onset of symptoms, but the number rapidly increased to 100.0% (total) [94.3% (IgM) and 79.8% (IgG)] by 15 days after onset[39]. Furthermore, IgM was not detected significantly earlier in time than IgG[40]. Serologic testing of infected individuals should be done no earlier than 1 week after symptom onset to minimize false negatives and preferably should be done at least 2 weeks after onset when most should have developed antibodies. Positive serology tests should be interpreted in context of the individual's course of infection. For instance, an individual with a positive antibody test 7 days after symptom onset may still have an acute infection and have a higher probability of transmitting the virus to others compared to one with a positive test 21 days after symptom onset. Clinicians should also consider the possibility that certain individuals (i.e. immunocompromised) may not mount any antibody response.

Self-collection

Most PCR swabs are taken by a trained collector. However, self-collection has logistical advantages and greater ease of collection may encourage more testing. As a result, some providers and patients may prefer self-collection of nasopharyngeal swabs. Little data exist on the relative validity of test results from self-collection versus collection by a trained collector. In a small study, supervised self-collection performed about as well as provider collection[41], though another study found lower sensitivity for self-collected nasopharyngeal samples but high sensitivity for self-collected saliva samples[42]. These findings are consistent with an earlier meta-analysis of influenza PCR sample found that sensitivity was 13% lower for self-collection, which uses similar technology as for SARS-CoV-2[43].

Testing Performance

The performance of commercial serological tests, especially for rapid antibody tests, varies widely. A meta-analysis evaluating rapid as well as ELISA tests revealed that pooled sensitivity and specificity were 82% and 97%, respectively, for IgM antibody tests and 97% and 98% for IgG antibody tests. The false negative rates for antibody tests also ranged from 10-44%[44]. Furthermore, a recent study evaluating 10 lateral flow assays found that sensitivity ranged from 81 to 100% (higher with samples taken 20 days after symptom onset) and specificity ranged from 84 to 100%. Detecting both IgM and IgG together increased sensitivity as well, suggesting combined antibody detection may aid in improving detection rates of serological tests[45]. With such varying statistics, clinicians cannot rule out COVID-19 in patients with a negative result.

Currently, there is conflicting data regarding cross-reactivity of antibody tests. One study evaluating the VivaDiag COVID-19 IgM and IgG Rapid Test found no cross-reactivity in 10 subjects with previous coronavirus infection[46]. Meanwhile, cross-reaction of sera from COVID-19 patients with SARS-CoV assays, including ELISA and immunofluorescence assay, was observed, possibly due to both viruses using the same receptor to enter host cells[47]. Thus, cross-reaction while using antibody tests, although unlikely, may be a possibility to consider when interpreting a positive antibody test result.

Application to practice: Clinical Scenarios

Because the indications for testing, the types of tests available, and the context in which tests are performed vary so widely, it is challenging for physicians to interpret the results of tests that have been performed as well as on how to advise their patients on whether and how to be tested. To illustrate these concerns this section presents three scenarios with different individual motivations for testing, use of different tests, and their application. In each of these scenarios, interpretation of a positive or negative test result should incorporate the pretest probability of infection, which is based on both general prevalence in the population and the patient's specific

risks; test sensitivity and specificity; and if applicable, the timing of the test relative to symptom onset and/or resolution.

Scenario 1 – Low pretest probability (LFA)

A healthy, young professional who lives alone in Vermont is required by his employer to get a COVID antibody test prior to returning to work in person. The individual does not work in health care and had no symptoms suggestive of COVID or known exposure. Given the place of residence and lack of symptoms, pretest probability or likelihood of having SARS-CoV2 infection is very low, perhaps 1 in 1000. In a population of 10,000, 10 would have the virus and 9,990 would not. The individual is offered a point-of-care antibody test, a lateral flow assay that detects both IgM and IgG antibodies. Suppose the sensitivity and specificity of the test are 90% and 95%, respectively. If all 10,000 individuals were to be tested with this antibody test, there would be 9 true positives, 500 false positives, 1 false negative and 9,490 true negatives. Since the negative predictive value is near 100%, a negative test result can be trusted with near certainty. A positive test result, given the low pretest probability and 1.8% positive predictive value, is most likely a false positive. As of September 2020, CDC recommends that serologic tests should not be used to diagnose COVID or make decisions on whether an individual can return to the workplace[48]. Anyone who does test positive for SARS-CoV2 antibodies should continue to wear a mask in public, avoid close social contact, and wash hands frequently, since it is uncertain if antibodies protect against repeat infection or prevent viral transmission.

Scenario 2 – Intermediate pretest probability (RT-PCR initially negative)

In this scenario (Table 2, Panel b), an older patient with COPD presents with shortness of breath and fever. Alternate diagnoses of COPD exacerbation, possibly with pneumonia, are as likely as COVID-19. His symptoms began a few days ago when he was at home. Although he visits the local grocery store once per week, he has not come in contact with anyone who tested positive. Suppose his pretest probability is 40%, as symptoms may be due to either his COPD or COVID-19. His upper respiratory tract is swabbed for an RT-PCR test and the result comes back negative. Assuming the test has sensitivity of 85% and specificity of 90%, there is 90.5% likelihood that he actually does not have an infection. A negative result does not, however, rule out infection with near certainty. Interpretation can be further complicated by the type of test used. For example, suppose the older patient had a week of symptoms and is inappropriately offered serological testing. A lateral flow assay, the same as one used in scenario 1, yields a negative test result. However, due to being tested within 7 days after symptom onset, the sensitivity is significantly lower (60%). Using the 60% pretest probability again, the positive and negative predictive values fall to 75% and 53.8%, respectively. A negative result in this case should be interpreted with caution and acknowledgement that infection cannot be ruled out.

Scenario 3 – High pretest probability (RT-PCR)

A healthy 20 year-old female comes into the student health clinic because one of the residents of her college dormitory was diagnosed with COVID yesterday after having had a dry cough and fever for a week.. She herself began having a dry cough for the past 3 days and a fever of 101.5 degrees Fahrenheit last night. A rapid molecular SARS-CoV2 test is offered. Given her symptoms and close direct contact with an individual who tested positive, her pretest probability is very high. The test comes back as negative a day later. Suppose the test has a sensitivity of 90% and specificity of 95% and her pretest probability is 80%. The predictive value for a positive test result would be 98.6% and a negative test result would be 70.4%. In other words, a positive test result can be trusted with high certainty, but a negative result could easily be a false negative. In this case, the symptomatic student should be self-isolated on campus despite testing negative. In accordance with CDC guidelines, isolation should be continued until at least 10 days have passed since symptom onset and at least 24 hours have passed since resolution of symptoms to minimize the risk of transmission of SARS-CoV2 to other students on campus[49].

Considerations for advising patients

With each passing day, more, and different types of tests become available, some less sensitive and specific than those currently available, but with lower cost and shorter turnaround times[50]. The indications for testing, and national and state goals for testing, also are constantly changing. Therefore, rather than make recommendations about specific tests and situations, our analysis focuses on test interpretation relevant to clinical practice.

Given the challenges in test interpretation summarized here, the first physicians must address with their patients is whether to be tested at all. For individuals with symptoms of COVID-19 or who have been identified as a positive contact should definitely be tested. So too for those whose employers or universities conduct a comprehensive testing program. However, if test results are so delayed that they are no longer actionable, as was the case in some parts of the United States in the summer of 2020,[51, 52] there is little value in testing. Indeed, patients in a low prevalence area of the country might get a false sense of assurance from a negative result of a low sensitivity test conducted at the wrong time. Such patients also run the unnecessary risk of having to quarantine based on a false positive result. Beyond this, the question of whether to test at all is complex, but physicians should consider what they and their patients should do differently if the test were positive or negative, as well as the likelihood and consequences of false negatives and positives.

If a test is conducted, clinicians should keep in mind several key considerations regarding the motivations for testing, test characteristics, and application of those characteristics in interpreting test results.

First, many factors shape who seeks a test and when. Specifically, policy pressures created by changing population norms for testing, organizational pressures created by hospitals, universities or employers setting guidelines for testing, and individual pressures created through social

interactions and perceptions, can increase testing demand and expectations. These pressures may result in peaks and valleys of when patients seek testing, and how they rely upon a clinician to interpret their test results such as advising on whether they can attend a certain event after a negative diagnostic test or go back to work after an antibody test. These pressures may also shape the specific test an individual desires or seeks. For example, if the motivation for testing is a rapid result to participate in activity, an antigen test that can deliver rapid results is more desirable to the individual; in contrast if the motivation is to comply with the needs of an employer, timing of results and sensitivity of results may be a higher priority. This suffices to say, which test is used is related to the testing motivations.

Second, testing performance depends on the balance between testing characteristics and sensitivity and specificity; i.e. is a false positive or false negative a better outcome when needing to make population decisions. Because most tests err on the side of having high sensitivity and lower specificity (i.e. there will be more false positives than false negatives), clinicians must be prepared to discuss these results with patients and contextualize an individual's result within the broader realm of the purposes testing can serve. While clinicians are often trained to think mainly about positive predictive value, negative predictive value is equally important for an infectious disease because diminished NPV means a higher percentage of people with negative tests are infectious. Patients should be reminded that risk reduction behaviors remain prudent after a negative test both because of the risk of a false negative and because infection can occur subsequent to the test.

Third, test results must be interpreted in the geographic and clinical context in which the test was administered. In particular, as the clinical scenarios illustrate, the reason a patient seeks testing is often a strong indicator of the pre-test probability of infection, and thus how to interpret test results. In addition, the level of population spread of the virus and the timing of testing will play critical roles in the positive or negative predictive value of the test. Clinicians should counsel patients that the value of an individual test result may vary as community spread of COVID-19 recedes or later resurges.

Table 1. Theoretical explanation of individual motivation for seeking a test based on the Health Belief Model (Ref 8)

Constructs	Definition	Application to testing motivation
Perceived Susceptibility	Belief about the likelihood of getting a disease or condition	Perception of COVID-19 being an issue in the individual's geographic area
Perceived severity	Belief about the seriousness of contracting a condition or of leaving it untreated, including physical consequences and social consequences	Perception the individual is at risk for COVID-19 given age and other demographics
Perceived benefits	Beliefs about positive features or advantages of a recommended action to reduce threat	Perception that a test provides beneficial information or is a "treatment" unto itself
Perceived barriers	Beliefs about negative features or of a recommended action to reduce threat	Perception that test is not useful or painful to get
Cue to action	Internal or external markers stimulus	Media coverage of testing; knowing others who have been tested; getting sick
Self-efficacy	The conviction that one can successfully execute a behavior	Ability to go and get a test

Table 2. Test performance in three hypothetical clinical scenarios

Panel a: Scenario 1 – Low pretest probability (LFA)

	Dx	No Dx	PPV	NPV
Prevalence 0.1%, Sn 90, Sp 95				
(+)	9	500	1.8%	99.9%
(-)	1	9490		
Prevalence 0.2%, Sn 90, Sp 95				
(+)	18	499	3.5%	100.0%
(-)	2	9,481		

Panel b: Scenario 2 – Intermediate pretest probability (RT-PCR initially negative)

	Dx	No Dx	PPV	NPV
Prevalence 40%, Sn 85%, Sp 95%				
(+)	34	3	91.9%	90.5%
(-)	6	57		
Prevalence 60%, Sn 85%, Sp 95%				
(+)	51	2	96.2%	80.9%
(-)	9	38		

Panel c: Scenario 3 – High pretest probability (RT-PCR)

	Dx	No Dx	PPV	NPV
Prevalence 80%, Sn 90%, Sp 95%				
(+)	72	1	98.6%	70.4%
(-)	8	19		
Prevalence 80%, Sn 80%, Sp 95%				
(+)	64	1	98.4%	54.3%
(-)	16	19		

Notes: these tables describe the distribution of 10,000 (panel a) or 100 patients (panels b and c) with and without the disease (COVID-19) compared to with positive and negative test results

- sensitivity (Se), the proportion of infected individuals who test positive
- specificity (Sp), the proportion of uninfected individuals who test negative
- Dx = patient has disease; no Dx = patient does not have disease
- (+) = test result is positive; (-) = test result is negative
- positive predictive value (PPV), the probability that a positive test result actually means one has COVID-19
- negative predictive value (NPV), the probability that a negative test result actually means one does not have COVID-19

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