

COVID-19 Diagnostic Testing

Reverse transcription-polymerase chain reaction (RT-PCR) testing for COVID-19 detects RNA from SARS coronavirus 2 (SARS-CoV-2) and is the primary test used for diagnosis of acute infection. RT-PCR is considered the gold standard for respiratory virus testing, with very high analytical sensitivity (the ability to detect virus from known positive and negative controls in the lab, generally >95% over numerous studies)¹. Limits of detection may vary somewhat between different test methods, but all FDA-Emergency Use Authorization (EUA)-approved tests for SARS-CoV-2 are expected to detect levels of virus that are present during acute symptomatic infection.

Clinical sensitivity of RT-PCR varies by site of sampling, likely due to variation in quality of sampling technique, time of sampling with respect to disease course (viral titers are highest early in infection^{2,7}), and variation in the distribution of virus in the upper versus lower respiratory tract. Our understanding of clinical sensitivity of RT-PCR is based on a) prior studies using RT-PCR to detect other respiratory viruses¹, and b) limited data on SARS-CoV-2⁴. Prior studies of respiratory viruses have found that sampling by nasopharyngeal (NP) swab may be more sensitive than oropharyngeal (OP) swab sampling, and that a combination of NP + OP may increase sensitivity, although variation by virus was observed⁴⁻⁵. Two limited studies of SARS-CoV-2 have compared percent test positivity based on sampling site over the disease course but were not done in a way that allowed accurate calculation of sensitivity. The larger study (213 patients, not-yet peer reviewed) found a decrease in viral detection over time, with test positivity in the first 14 days after symptom onset somewhat higher in NP swabs (72%) versus OP swabs, and with lower respiratory samples remaining positive in severe disease². The smaller study (9 patients) found high viral loads in pharyngeal samples during the first week of symptoms, with 100% test positivity during the first five days of symptoms and 46% test positivity after the first five days, independent of swab type⁷.

While more information is needed and there likely is individual variation in the time course of viral shedding, existing studies suggest that clinical sensitivity is likely very high during the early acute, symptomatic phase of infection, but lower during later stages of infection when viral titers may be low. No data on test sensitivity in asymptomatic infected individuals exists. Additional studies suggest that older patients have higher viral loads and that sputum and lower respiratory specimens (endotracheal aspirate) have high viral loads in patients with severe pulmonary disease^{7,8}. A recent study (not yet peer-reviewed) suggests that patient self-sampling of the anterior nares or mid turbinate nares may have comparable sensitivity to that of a clinician administered NP swab¹⁰.

What does a negative RT-PCR test mean? The negative predictive value (NPV) is the probability that a person with a negative test does not have infection and differs depending on the prevalence of disease in the population. In asymptomatic patients, based on existing data,¹¹ the prevalence of active SARS-CoV-2 viral infection in the Bay Area and in the U.S. is estimated to be approximately 1% at any given time. Given that the estimated prevalence of asymptomatic patients in the Bay Area is very low, the negative predictive value for testing an asymptomatic patient prior to surgery is very high. For example, if the prevalence is assumed to be 1% and the sensitivity/specificity of a NP+OP swab test is estimated at 80%/98% (sensitivity estimate based on sampling at an unknown time over the entire course of infection), then the negative predictive value of the test is 99.8%. In symptomatic patients or those with known exposures, the prevalence may be 10% or higher. In this case, the negative predictive value is estimated to be 97.2%.

Main points

1. RT-PCR is the diagnostic test of choice
 2. Sensitivity is high early in symptomatic infection but decreases over time
 3. Recommended specimens:
 - Combined NP swab + OP swab
 - Combined Mid turbinate + OP swab
 - Tracheal aspirate (intubated patients)
 4. Estimated NPV of a single PCR test in a patient in the Bay Area currently:
 - asymptomatic patient: 99.8%
 - symptomatic patient: 97.8%
 5. When to use serology:
 - patients with negative PCR testing strongly suspected to have COVID-19
 - documentation of seropositivity for plasma donation or vaccine studies
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Serologic Testing detects antibodies to SARS-CoV-2 produced by the patient. Antibodies may not reach levels high enough to detect until 7-14 days after infection⁷, and may never be detected in patients with impaired immunity. There is significant variability in commercial serologic assays and some point-of-care assays may have high false positivity rates due to cross-reactivity with 4 common seasonal coronaviruses that circulate widely in the United States. We don't yet know how antibody detection using certain assays correlates with protective immunity. The primary uses of serologic testing at UCSF are: 1) to improve sensitivity for COVID-19 diagnosis in patients with a high suspicion of COVID-19 but negative RT-PCR testing presenting more than a week into infection and 2) to document antibody response in patients planning to donate convalescent plasma or participating in vaccine trials.

Clinical Evaluation Guide for Diagnosis and Testing of COVID-19:

<https://infectioncontrol.ucsfmedicalcenter.org/sites/g/files/tkssra4681/f/COVID-19%20Clinical%20Evaluation%20Guide.pdf>

UCSF Inpatient Adult COVID-19 Interim Management Guidelines:

https://infectioncontrol.ucsfmedicalcenter.org/sites/g/files/tkssra4681/f/UCSF_Adult_COVID_draft_management_guidelines.pdf

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