

REVIEW

An evolving approach to the laboratory assessment of COVID-19

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Email: luhongzhou@fudan.edu.cnYi-Wei Tang, Cepheid, Danaher Diagnostic Platform, 518 Fuquan North Road, 200325 Shanghai, China.
Email: yi-wei.tang@cepheid.com**Abstract**

As the 2019 novel coronavirus disease (COVID-19) outbreak has evolved in each country, the approach to the laboratory assessment of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection has had to evolve as well. This review addresses the evolving approach to the laboratory assessment of COVID-19 and discusses how algorithms for testing have been driven, in part, by the demand for testing overwhelming the capacity to accomplish such testing. This review focused on testing in the USA, as this testing is evolving, whereas in China and other countries such as South Korea testing is widely available and includes both molecular testing for SARS-CoV-2 as well as serological testing using both enzyme-linked immunosorbent assay methodology and lateral flow immunoassay methodology. Although commercial testing systems are becoming available, there will likely be insufficient numbers of such tests due to high demand. Serological testing will be the next testing issue as the COVID-19 begins to subside. This will allow immunity testing as well as will allow the parameters of the COVID-19 outbreak to be defined.

KEYWORDS

assessment, COVID-19, molecular testing, serology

1 | INTRODUCTION

As the world continues to cope with the 2019 novel coronavirus disease (COVID-19) pandemic,¹⁻⁵ testing methods and algorithms for their use in the assessment of COVID-19 are rapidly evolving. This update will specifically address those testing methods and algorithms for the assessment of COVID-19 that are currently in use or will be available for use in the near future. This review focused on testing in the USA, as this testing is evolving, whereas in China and other countries such as South Korea testing is widely available and includes both molecular testing for severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) as well as serological testing using both enzyme-linked immunosorbent assay (ELISA) methodology and lateral flow immunoassay methodology. The limitations of current testing methods will be discussed, as these limitations have led to the use of testing algorithms for COVID-19 testing. In addition, serological testing for the assessment of immunity to COVID-19 will be discussed as such serological results may be useful for determining who can return to work in critical occupations.

Rapid, commercial COVID-19 testing methods as well as algorithms for their use are being developed and implemented throughout the world. These will assist in the control and final resolution of this outbreak. As the COVID-19 public-health emergency rapidly evolves country by country, the responses in each country must meet the emerging needs. But a common factor for every country is the need for wide-spread testing at the level of small communities.⁶ As the COVID-19 outbreak begins to subside, serological testing will be needed to determine who is likely to be immune and thus might be able to return to work, particularly if such work is a critical occupation. Serological testing will also be needed to determine the epidemiological characteristics such as the true case fatality rate.

1.1 | Specific types of molecular testing

Wide availability at the community level for commercial molecular testing instruments that can use approved SARS-CoV-2 test kits will be

absolutely critical for controlling this COVID-19 outbreak. As many hospitals and clinics already are using such instruments, they would only need to verify the SARS-CoV-2 testing on their instruments to be able to offer such testing at the local community level. In the USA, this will require the Food and Drug Administration (FDA) approval for “emergency use authorization” (EUA) for in vitro diagnostic use tests. At the time of writing, 39 kits, most of them are commercial except for the one from the US Centers for Disease Control and Prevention (CDC) and the NY State of Health, received EUA clearance (<https://www.fda.gov/medical-devices/emergency-situations-medical-devices/emergency-use-authorizations#coronavirus2019>. Accessed 18 April 2020). However, the demand for these commercial COVID-19 devices could easily exceed the capability of their respective manufacturers to supply a large initial demand.

1.1.1 | Random-access, integrated, and sealed devices

Many of these random-access devices are real-time reverse-transcription polymerase chain reaction (RT-PCR)-based procedures which do include a separate nucleic acid extraction procedure before the procedure of simultaneous nucleic acid amplification and detection. However, several commercial testing instruments now offer “sealed” systems that integrate nucleic acid extraction, amplification, and detection; once the clinical sample in universal transport medium is loaded into the instrument's cartridge in a biosafety cabinet, the cartridge is “sealed.”⁷⁻¹⁰ At the time of writing, Xpert Xpress SARS-CoV-2 test (Cepheid, Sunnyvale, CA), Accula SARS-CoV-2 Test (Mesa Biotech, San Diego, CA), and ID NOW COVID-19 (Abbott, Scarborough, ME) have been granted EUA CLIA-waive category by FDA. There are certain types of high priority testing algorithms for SARS-CoV-2 testing; these include testing of health-care workers¹¹ as health-care workers are at high risk and, if undiagnosed, could infect their patients. Random-access point of care instruments would be very useful for such high priority testing algorithms in the clinical microbiology laboratory, even if a high-throughput instrument for COVID-19 testing is also being used.

1.1.2 | Large scale, high-throughput molecular testing instruments

There are multiple large scale, high-throughput molecular testing instruments available.¹² The best known include the Roche 6800/8800, NeuMoDx Molecular Instrument, Luminex NxTAG, and Hologic Panther Fusion Instruments.¹³⁻¹⁶ These are larger platforms that can process and test between several hundred and several thousand tests per day. Because such large scale, high-throughput molecular testing instruments use batched testing, this testing takes longer. Some medical centers are using random-access point of care COVID-19 testing kits for priority testing while using the slower large scale, high-throughput COVID-19 testing platforms for epidemiological purposes.

1.2 | Testing consumables

Access to COVID-19 testing in many countries, including the USA, continues to be inadequate, which greatly impedes the ability of clinicians and public-health authorities to accurately determine and track the prevalence and transmissibility of this outbreak. Moreover, inadequate testing capabilities and/or testing delays also may hamper the clinicians' ability to appropriately identify, isolate, and treat these patients. One factor that has already been noted to limit the ability of COVID-19 testing is the scarcity of many testing consumables. A number of these scarcities will be discussed in the following subsections.

1.2.1 | Swabs with universal/viral transport medium

The molecular diagnosis of COVID-19 typically begins with a nasopharyngeal swab (NP) or an oropharyngeal swab (OP) collected and transported to the laboratory in universal/viral transport medium.¹⁷⁻²⁰ It is hard to imagine that these swabs, viral transport media, screw-cap tubes, plastic bags to transport the specimens, cold packs to cool the specimen, and styrofoam containers would become scarce, but this is happening in the USA as COVID-19 testing efforts are increasing. Several medical centers have resorted to making their own viral transport medium to meet this need. Saline, phosphate buffered saline, and minimum essential medium were even evaluated as potential alternatives to viral transport media for SARS-CoV-2 testing.²¹ Needless to say, this outbreak was not anticipated, and manufacturers of these specimen collection items require time to increase their production. Readers are suggested to follow CLSI M40 (Quality Control of Microbiological Transport Systems; Approved Standard) to validate different collection matrixes.

A recognized limitation of the use of OP or NP swabs for collecting upper respiratory tract specimens for COVID-19 molecular testing is that false-negative results of these initial real-time RT-PCR assays are known to occur^{22,23} (<https://doi.org/10.1101/2020.04.16.20066787>. Accessed 23 April 2020). One approach to this issue with false-positive results from OP or NP swabs in both China and the USA has been the use of self-collected saliva²⁴ (<https://doi.org/10.1101/2020.04.11.20062372>. Accessed 23 April 2020). Indeed, in one such study, the self-collected saliva was more sensitive for SARS-CoV-2 detection than was the NP swab (<https://doi.org/10.1101/2020.04.16.20067835>. Accessed 23 April 2020).

1.2.2 | Extraction mix reagents

For these commercial devices in which nucleic acid extraction kits are not provided, the users need to choose and purchase separate reagents. Extraction mix reagents also have become scarce as COVID-19 testing demands increase. Currently, commercially available nucleic acid extraction kits based on magnetic bead binding are widely used and include bioMerieux easyMAG or EMAG^{13,25} or QIAGEN

EZ1/QIAAsymphony.^{26,27} The advantage to use these extraction kits is laboratory safety as the buffers included in extraction systems do contain guanidinium/detergents and are able to inactivate SARS-CoV-2.²⁸⁻³⁰ However, these extraction mix reagents have become difficult to obtain, thus limiting COVID-19 testing.

1.2.3 | Reverse transcriptase and DNA polymerase mix reagents

For these laboratories have validated and implemented laboratory-developed testing for COVID-19, reverse transcriptase is combined with DNA polymerase to amplify the positive-sense RNA coronavirus; these DNA polymerase mix reagents also have become scarce as they have been consumed in early COVID-19 testing. Some laboratories have had to validate three different DNA polymerases mix reagents and three different extraction mix reagents so that their testing methods could easily shift and use whatever reagents are currently available.

1.3 | Testing algorithms and interpretation

Because availability of COVID-19 diagnostic testing is limited, the Infectious Diseases Society of America (IDSA) had developed a testing algorithm to help clinicians test wisely as well as for the laboratory reporting of these results (<https://www.idsociety.org/globalassets/idsa/public-health/covid-19-idsa-testing-intro.pdf>. Accessed 26 March 2020). Importantly, the IDSA has recommended that the CDC should publicly disclose the number of

cases tested in any one location or state to better gauge the significance of the number of positive results. The number of positive results in locations or states that have few or no reported cases may signify low or no prevalence of COVID-19 or may mean that such low-positive results are due to undertesting. This, in turn, will determine how aggressive testing needs to be in these locations or states. At the time of writing, the IDSA recommends a four-tier approach to COVID-19 diagnostic testing as shown in Table 1.

1.4 | Serological assays

1.4.1 | SARS-CoV-2 antigens

Members of the coronavirus family have four major structural proteins: the spike (S), membrane (M), envelope (E), and nucleocapsid (N) proteins. Two of these proteins appear to be important antigenic sites and thus have been utilized for the development of serological assays to detect COVID-19.^{22,31} The N protein of coronaviruses is a structural component of the helical nucleocapsid and has an important function in viral pathogenesis, replication, and RNA packaging. Antibodies to the N protein are frequently detected in COVID-19 patient³² (<https://doi.org/10.1101/2020.03.18.20038018>. Accessed 6 April 2020), suggesting that the N protein may be one of the immunodominant antigens in the early diagnosis of COVID-19.³³ The second antigenic site is the S protein and the S1 protein. The entry of coronavirus into host cells is accomplished by the transmembrane S glycoprotein that forms homotrimers protruding from the viral surface.³⁴

TABLE 1 IDSA four-tier approach to COVID-19 diagnostic testing

Tier level	Population
1	<ul style="list-style-type: none"> Critically ill patients receiving ICU level care with unexplained viral pneumoniae or respiratory failure, regardless of travel history or close contact with suspected or confirmed COVID-19 patients Any person, including health-care workers, with fever or signs/symptoms of a lower respiratory tract illness and close contact with a laboratory-confirmed COVID-19 patient within 14 d of symptom onset (including all residents at a long-term care facility that has a laboratory-confirmed COVID-19 case) Any person, including health-care workers, with fever or signs/symptoms of a lower respiratory tract illness and a history of travel within 14 d of symptom onset to geographical regions where sustained community transmission has been identified; (iv) individuals with fever or signs/symptoms of a lower respiratory tract illness who are also immunosuppressed (including patients with HIV), elderly, or have underlying chronic health conditions Individuals with fever or signs/symptoms of a lower respiratory tract illness who are critical to pandemic response, including health-care workers, public-health officials, and other essential leaders
2	<ul style="list-style-type: none"> Hospitalized (non-ICU) patients and long-term care residents with unexplained fever and signs/symptoms of a lower respiratory tract illness. The number of confirmed COVID-19 cases in the community should be considered. As testing becomes more widely available, routine testing of hospitalized patients may be important for infection prevention and management of discharge
3	<ul style="list-style-type: none"> Patients in outpatient settings who meet the criteria for influenza testing. This includes individuals with comorbid conditions including diabetes, COPD, congestive heart failure, age more than 50, immunocompromised hosts among others. Given limited available data, testing of pregnant women and symptomatic children with similar risk factors for complications is encouraged. The number of confirmed COVID-19 cases in the community should be considered
4	<ul style="list-style-type: none"> For community surveillance as directed by public-health and/or infectious diseases authorities

Abbreviations: COPD, chronic obstructive pulmonary disease; COVID-19, 2019 novel coronavirus disease; ICU, intensive care unit; IDSA, Infectious Diseases Society of America.

1.4.2 | Serological assay methods

The development of serological assays such as ELISA took a significant amount of time after the 2002 to 2003 SARS-CoV outbreak because creation of these assays was not a trivial process.³³ Recombinant antigens may allow serological assays for COVID-19 to be developed on a faster timeline. Several serological tests have been commercially available for COVID-19^{32,35} (<https://doi.org/10.1101/2020.03.17.20036954>; <https://doi.org/10.1101/2020.03.18.20038018>; <https://doi.org/10.1101/2020.03.17.20037713>. Accessed 6 April 2020). One of them, qSARS-CoV-2 IgG/IgM Rapid Test from Cellex Inc, received the US FDA EUA on 1 April 2020. This assay was granted emergency use in authorized laboratories for qualitative detection of IgM and IgG antibodies against SARS-CoV-2 in serum, plasma (EDTA or citrate), or venipuncture whole blood from individuals suspected of COVID-19 by their health-care provider (<https://www.fda.gov/media/136622/download>. Accessed 6 April 2020). At the time of writing, three additional ones, VITROS Immunodiagnostic Products Anti-SARS-CoV-2 Total Reagent Pack (Ortho Clinical Diagnostics, Rochester, NY), DPP COVID-19 IgM/IgG System (Chembio Diagnostic System, Medford, NY), and COVID-19 ELISA IgG Antibody Test (Mount Sinai Laboratory, New York, NY) have been granted EUA by FDA. Rapid antigen/antibody lateral flow immunoassays (LFIA) already have been developed for the diagnosis of COVID-19; additional assays will undoubtedly follow these.^{36,37} However, the early promise of LFIA devices has been questioned following concerns about sensitivity and specificity. A recent study parallelly compared one ELISA and nine different commercially available LFIA devices for detection of SARS-CoV-2-specific IgM and IgG antibodies revealing the performance of current LFIA devices was inadequate for most individual patient applications (<https://doi.org/10.1101/2020.04.15.20066407>. Accessed 23 April 2020). A pseudovirus-based neutralization assay for SARS-CoV-2 was developed and validated.¹⁰

1.4.3 | Role of serological assays

Such serological assays are critically important to determine the seroprevalence in a given population and to define previous exposure

as well as to provide information about asymptomatic patients that may have played a large role in transmitting COVID-19 (<https://doi.org/10.1101/2020.03.18.20037994>. Accessed 6 April 2020).^{38,39} Serology can be used to contact tracing, which is especially useful during current shortage of molecular assays. Serology results can be used in COVID-19 case confirmation when viral RNA-negative patients presenting late in the illness.⁴⁰ Beyond diagnosis, serology results may be used to guide return-to-work decision.⁴¹

However, antibody detection measures past exposure to SARS-CoV-2 and another production is host dependent and takes time. As a natural delay, antibody testing is not useful in the setting of an acute illness.^{22,23} A recent study validated the VivaDiag IgM/IgG Rapid Test³⁷ in acute patients referring to emergency room department in Italy. The sensitivity and specificity were 18.4% and 91.7%, indicating the serology assay should be avoided for triage of patients with suspected COVID-19.²⁶ Possible explanations included the low antibody titers or delayed humoral responses in patients with SARS-CoV-2 infections.⁴² A recent study revealed that the presence of anti-SARS-CoV-2 IgG did not start till the illness day 11 and postexposure 18 to 21 days.⁴³ Although unlikely to be very useful for the diagnosis of acute COVID-19, serology, especially ELISA-based ones, will be very useful for defining immunity as well as better characterizing certain parameters such as the true fatality rate. Result interpretation in the clinical setting in combination of molecular results is listed in Table 2.

2 | SUMMARY

As the COVID-10 outbreak has evolved, so has the approach to SARS-CoV-2 testing. Availability of molecular testing for COVID-19 has been limited in many areas due to a lack of consumable reagents. This has required testing algorithms. Fortunately, random-access point-of-care devices will soon be able to bring molecular testing to small communities. Initial availability of these kits may be limited due to a high demand. As these instruments become available, this capability combined with ELISA-based serological testing for immunity will further assist with the resolution of this outbreak.

TABLE 2 General molecular and serology test result interpretation in COVID-19

RNA	IgM	IgG	Interpretation
+	-	-	Patient in the 2-wk period before immune response
+	+	-	Patient in early infection
+	-	+	Patient in mid to late infections; confirmation if IgG titer in convalescence is four times higher than acute phase
+	+	+	Patient in active infection with decent immune response
-	+	-	Patient has active infection with a false-negative RNA assay
-	-	+	Patient with previous infection; virus has been cleared
-	+	+	Patient with recent infection and in convalescence; virus has been cleared; active infection with false-negative RNA assay

Abbreviation: COVID-19, 2019 novel coronavirus disease.

CONFLICT OF INTERESTS

YWT is an employee of Cepheid, the commercial manufacturer of the Xpert Xpress SARS-CoV-2 assay.

AUTHOR CONTRIBUTIONS

HL and YWT assisted in concept forming; HL, CWS, and YWT contributed in content development; CWS and YWT helped in writing the article.

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REFERENCES

- Bedford J, Enria D, Giesecke J, et al. COVID-19: towards controlling of a pandemic. *Lancet*. 2020;395(10229):1015-1018. [https://doi.org/10.1016/S0140-6736\(20\)30673-5](https://doi.org/10.1016/S0140-6736(20)30673-5)
- Hick JL, Biddinger PD. Novel coronavirus and old lessons - preparing the health system for the pandemic [published online ahead of print March 25, 2020]. *N Engl J Med*. 2020. <https://doi.org/10.1056/NEJMp2005118>
- Lu H, Stratton CW, Tang YW. The Wuhan SARS-CoV-2-what's next for China. *J Med Virol*. 2020;1(10):25738-547.
- Lu H, Stratton CW, Tang YW. Outbreak of pneumonia of unknown etiology in Wuhan, China: the mystery and the miracle. *J Med Virol*. 2020;92(4):401-402. <https://doi.org/10.1002/jmv.25678>
- Rothan HA, Byrareddy SN. The epidemiology and pathogenesis of coronavirus disease (COVID-19) outbreak. *J Autoimmun*. 2020;109:102433. <https://doi.org/10.1016/j.jaut.2020.102433>
- Rubin EJ, Baden LR, Morrissey S. Audio interview: emerging tools in the fight against Covid-19. *N Engl J Med*. 2020;382(15):e39. <https://doi.org/10.1056/NEJMe2009651>
- Chandler DP, Bryant L, Griesemer SB, et al. Integrated amplification microarrays for infectious disease diagnostics. *Microarrays (Basel)*. 2012;1(3):107-124. <https://doi.org/10.3390/microarrays1030107>
- Babady NE, Mead P, Stiles J, et al. Comparison of the Luminex xTAG RVP Fast assay and the Idaho Technology FilmArray RP assay for detection of respiratory viruses in pediatric patients at a cancer hospital. *J Clin Microbiol*. 2012;50(7):2282-2288. <https://doi.org/10.1128/JCM.06186-11>
- Ling L, Kaplan SE, Lopez JC, Stiles J, Lu X, Tang YW. Parallel validation of three molecular devices for simultaneous detection and identification of influenza A and B and respiratory syncytial viruses. *J Clin Microbiol*. 2018;56(3):e01691-17. <https://doi.org/10.1128/JCM.01691-17>
- Nie J, Li Q, Wu J, et al. Establishment and validation of a pseudovirus neutralization assay for SARS-CoV-2. *Emerg Microbes Infect*. 2020;9(1):680-686. <https://doi.org/10.1080/22221751.2020.1743767>
- Ng K, Poon BH, Kiat Puar TH, et al. COVID-19 and the risk to health care workers: a case report [published online ahead of print March 16, 2020]. *Ann Intern Med*. 2020. <https://doi.org/10.7326/L20-0175>
- Pfefferle S, Reucher S, Norz D, Lutgehetmann M. Evaluation of a quantitative RT-PCR assay for the detection of the emerging coronavirus SARS-CoV-2 using a high throughput system. *Euro Surveill*. 2020;25(9):2000152. <https://doi.org/10.2807/1560-7917.ES.2020.2525.2809.2000152>
- Tang YW, Gonsalves S, Sun JY, et al. Clinical evaluation of the Luminex NxTAG respiratory pathogen panel. *J Clin Microbiol*. 2016;54(7):1912-1914. <https://doi.org/10.1128/JCM.00482-16>
- Wirdein M, Larrouy L, Mahjoub N, et al. Multicenter comparison of the new Cobas 6800 system with Cobas Ampliprep/Cobas TaqMan and Abbott RealTime for the quantification of HIV, HBV and HCV viral load. *J Clin Virol*. 2017;96:49-53. <https://doi.org/10.1016/j.jcv.2017.1009.1007>
- Emery CL, Relich RF, Davis TH, Young SA, Sims MD, Boyanton BL Jr. Multicenter evaluation of NeuMoDx group B *Streptococcus* assay on the NeuMoDx 288 molecular system. *J Clin Microbiol*. 2019;57(2):e01324-18. <https://doi.org/10.1128/JCM.01324-18>
- Banerjee D, Kanwar N, Hassan F, Essmyer C, Selvarangan R. Comparison of six sample-to-answer influenza A/B and respiratory syncytial virus nucleic acid amplification assays using respiratory specimens from children. *J Clin Microbiol*. 2018;56(11):e00930-18. <https://doi.org/10.1128/JCM.00930-18>
- Druce J, Garcia K, Tran T, Papadakis G, Birch C. Evaluation of swabs, transport media, and specimen transport conditions for optimal detection of viruses by PCR. *J Clin Microbiol*. 2012;50(3):1064-1065. <https://doi.org/10.1128/JCM.06551-11>
- Goff J, Rowe A, Brownstein JS, Chunara R. Surveillance of acute respiratory infections using community-submitted symptoms and specimens for molecular diagnostic testing. *PLoS Curr*. 2015;7. <https://doi.org/10.1371/currents.outbreaks.0371243baa7f3810ba1279e30b96d3b6>
- Kim C, Ahmed JA, Eidex RB, et al. Comparison of nasopharyngeal and oropharyngeal swabs for the diagnosis of eight respiratory viruses by real-time reverse transcription-PCR assays. *PLoS One*. 2011;6(6):e21610. <https://doi.org/10.1371/journal.pone.0021610>
- To KK, Lu L, Yip CC, et al. Additional molecular testing of saliva specimens improves the detection of respiratory viruses. *Emerg Microbes Infect*. 2017;6(6):e49-7. <https://doi.org/10.1038/emi.2017.1035>
- Rodino KG, Espy MJ, Buckwalter SP, et al. Evaluation of saline, phosphate buffered saline and minimum essential medium as potential alternatives to viral transport media for SARS-CoV-2 testing [published online ahead of print March 30, 2020]. *J Clin Microbiol*. 2020. <https://doi.org/10.1128/JCM.00590-20>
- Loeffelholz MJ, Tang YW. Laboratory diagnosis of emerging human coronavirus infections – the state of the art. *Emerg Microbes Infect*. 2020;9(1):747-756.
- Tang YW, Schmitz JE, Persing DH, Stratton CW. The laboratory diagnosis of COVID-19 infection: current issues and challenges [published online ahead of print April 03, 2020]. *J Clin Microbiol*. 2020. <https://doi.org/10.1128/JCM.00512-20>
- To KKW, Tsang OTY, Yip CCY, et al. Consistent detection of 2019 novel coronavirus in saliva [published online ahead of print February 12, 2020]. *Clin Infect Dis*. 2020. <https://doi.org/10.1093/cid/ciaa149>
- Hindiyeh M, Mor O, Pando R, et al. Comparison of the new fully automated extraction platform eMAG to the MagNA PURE 96 and the well-established easyMAG for detection of common human respiratory viruses. *PLoS One*. 2019;14(2):e0211079. <https://doi.org/10.1371/journal.pone.0211079>
- Cassaniti I, Novazzi F, Giardina F, et al. Performance of VivaDiagTM COVID-19 IgM/IgG rapid test is inadequate for diagnosis of COVID-19 in acute patients referring to emergency room department [published online ahead of print March 30, 2020]. *J Med Virol*. 2020. <https://doi.org/10.1002/jmv.25800>
- Yera H, Filisetti D, Bastien P, Ancelle T, Thulliez P, Delhaes L. Multicenter comparative evaluation of five commercial methods for toxoplasma DNA extraction from amniotic fluid. *J Clin Microbiol*. 2009;47(12):3881-3886. <https://doi.org/10.1128/JCM.01164-09>
- Blow JA, Dohm DJ, Negley DL, Mores CN. Virus inactivation by nucleic acid extraction reagents. *J Virol Methods*. 2004;119(2):195-198. <https://doi.org/10.1016/j.jviromet.2004.03.015>
- Burton JE, Easterbrook L, Pitman J, et al. The effect of a non-denaturing detergent and a guanidinium-based inactivation agent on the viability of Ebola virus in mock clinical serum samples. *J Virol Methods*. 2017;250:34-40. <https://doi.org/10.1016/j.jviromet.2017.09.020>

30. Kumar M, Mazur S, Ork BL, et al. Inactivation and safety testing of Middle East respiratory syndrome coronavirus. *J Virol Methods*. 2015; 223:13-18. <https://doi.org/10.1016/j.jviromet.2015.07.002>
31. Petherick A. Developing antibody tests for SARS-CoV-2. *Lancet*. 2020; 395(10230):1101-1102. [https://doi.org/10.1016/S0140-6736\(20\)30788-1](https://doi.org/10.1016/S0140-6736(20)30788-1)
32. Okba NMA, Müller MA, Li W, et al. Severe acute respiratory syndrome coronavirus 2-specific antibody responses in coronavirus disease 2019 patients. *Emerg Infect Dis*. 2020;26(7). <https://doi.org/10.3201/eid2607.200841>
33. Meyer B, Drosten C, Müller MA. Serological assays for emerging coronaviruses: challenges and pitfalls. *Virus Res*. 2014;194:175-183. <https://doi.org/10.1016/j.virusres.2014.03.018>
34. Tortorici MA, Veesler D. Structural insights into coronavirus entry. *Adv Virus Res*. 2019;105:93-116. <https://doi.org/10.1016/bs.aivir.2019.08.002>
35. Zhang W, Du RH, Li B, et al. Molecular and serological investigation of 2019-nCoV infected patients: implication of multiple shedding routes. *Emerg Microbes Infect*. 2020;9(1):386-389. <https://doi.org/10.1080/22221751.2020.1729071>
36. Kozula KM, Gallotta A. Lateral flow assays. *Essays Biochem*. 2016; 60(1):111-120. <https://doi.org/10.1042/EBC20150012>
37. Li Z, Yi Y, Luo X, et al. Development and clinical application of a rapid IgM-IgG combined antibody test for SARS-CoV-2 infection diagnosis [published online ahead of print February 27, 2020]. *J Med Virol*. 2020;(10). <https://doi.org/10.1002/jmv.25727>
38. Hu Z, Song C, Xu C, et al. Clinical characteristics of 24 asymptomatic infections with COVID-19 screened among close contacts in Nanjing, China. *Sci China: Life Sci*. 2020;63(5):706-711.
39. Li R, Pei S, Chen B, et al. Substantial undocumented infection facilitates the rapid dissemination of novel coronavirus (SARS-CoV2) [published online ahead of print March 16, 2020]. *Science*. 2020; eabb3221. <https://doi.org/10.1126/science.abb3221>
40. Zhao J, Yuan Q, Wang H, et al. Antibody responses to SARS-CoV-2 in patients of novel coronavirus disease 2019 [published online ahead of print March 28, 2020]. *Clin Infect Dis*. 2020;ciaa344. <https://doi.org/10.1093/cid/ciaa344>
41. Patel R, Babady E, Theel ES, et al. Report from the American Society for Microbiology COVID-19 International Summit, 23 March 2020: value of diagnostic testing for SARS-CoV-2/COVID-19. *mBio*. 2020; 11(2):e00722-20. <https://doi.org/10.1128/mBio.00722-20>
42. Wölfel R, Corman VM, Guggemos W, et al. Virological assessment of hospitalized patients with COVID-2019 [published online ahead of print April 1, 2020]. *Nature*. 2020. <https://doi.org/10.1038/s41586-020-2196-x>
43. Lee NY, Li CW, Tsai HP, et al. A case of COVID-19 and pneumonia returning from Macau in Taiwan: clinical course and anti-SARS-CoV-2 IgG dynamic [published online ahead of print March 10, 2016]. *J Microbiol Immunol Infect*. 2020;S1684. <https://doi.org/10.1016/j.jmii.2020.03.003>

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