CURRENT DIAGNOSTIC AND THERAPEUTIC STRATEGIES FOR COVID-19- A REVIEW

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Abstract:
The outbreak and spread of the novel coronavirus disease 2019 (COVID-19) with pandemic features, which is caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), have greatly threatened global public health. Given the perniciousness of COVID-19 pandemic, acquiring a deeper understanding of this viral illness is critical for the development of new vaccines and therapeutic options. In this review, we introduce the systematic evolution of coronaviruses and the structural characteristics of SARS-CoV-2. We also summarize the current diagnostic tools and therapeutic strategies for COVID-19.

Keywords: Diagnosis; Treatment; COVID-19; Coronavirus; SARS-CoV-2.

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INTRODUCTION:
The outbreak and spread of novel coronavirus disease 2019 (COVID-19) pose the most serious threat to global public health. According to the World Health Organization (WHO), this disease was designated as a “public health emergency of international concern” on January 30, 2020. The corresponding virus was officially designated as 2019-nCoV by the Chinese Centre for Disease Control and Prevention, and later revised to severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) due to a good sequence homology towards SARS-CoV genome. Genomic sequencing demonstrated that SARS-CoV-2 was 96.2% identical to a bat coronavirus and shared a 79.5% sequence similarity to SARS-CoV, indicating that this virus is most likely to originate from bats [1].

COVID-19 has become a major concern globally, with over 70 million confirmed cases and 1.6 million lives lost as of December 15, 2020. Moreover, millions of people’s lives are at great risk because COVID-19 can be easily transmitted through direct contact with an infected person or indirect exposure to contaminated items. Health officials have approved some evidences of a transmission chain with four “generations”, and proposed a sustained human-to-human transmission. It has been speculated that a person infected with SARS-CoV-2 may infect more than three healthy individuals (the average reproductive number is approximately 3.28). COVID-19 also has a major impact on the global economy and affects the day-to-day life of individuals. Businesses have shut down; schools have been suspended and stores have closed as a result of this disease. Therefore, COVID-19 will likely continue to become a global health challenge and have far-reaching economic impacts if no control measures are in place [2].

The development of diagnostics and therapeutics is in progress, but there is currently no vaccine or antiviral drug for the treatment of COVID-19. Diagnostic tools have a crucial role in the battle against COVID-19, which allows for the rapid implementation of control measures to suppress SARS-CoV-2 transmission via case identification, contact tracing, and isolation of positive cases [3]. The mainstay for the clinical management of COVID-19 patients is symptomatic treatment instead of curative treatment, where critically ill patients are treated with ventilatory support in the intensive care unit [4]. Other common treatment strategies include antiviral therapy, antibiotic therapy, and immunomodulating therapy [5]. In this review, we first introduce the systematic evolution of coronaviruses (CoVs) and the structural characteristics of SARS-CoV-2. We then focus on known diagnostic tools and strategies for detecting SARS-CoV-2 in order to suppress its spread. Finally, we discuss current therapeutic methods, potential drugs, and vaccines. The overall intent of this review is to provide diagnostic and treatment strategies for dealing with COVID-19 pandemic through a summary of current findings [6].

Fig No.1: Schematic representation of transmission, treatment and vaccines of Covid-19
Similarities and Differences between SARS and COVID-19

Similarities between SARS and COVID-19
Like the more advanced cases of COVID-19, SARS manifested as a rapidly progressing viral pneumonia. The primary mode of transmission of SARS and COVID-19 appears to be via infectious respiratory droplets dispersed from the mucous membranes (Table 1). SARS-CoV and SARS-CoV-2 are reported to have similar stability and decay rate in aerosols and on several surfaces. It has been demonstrated that both can survive for up to 3 days on plastic and up to 2 days on stainless steel, with similar decay profiles of the virus titre on each surface. The median incubation period, which is the time from the initial exposure until the onset of symptoms, appears to be around 4–7 days, and the maximum incubation period could be up to 14 days for both SARS and COVID-19[6].

Table 1. Similarities and Differences between SARS and COVID-19

|                          | SARS                  | COVID-19              |
|--------------------------|-----------------------|-----------------------|
| **Pre-Transmissibility** | NO                    | YES                   |
| **Mild Case Transmissibility** | NO                | YES                   |
| **Reproduction Number (R_0)** | 1.7–1.9 (WHO)  | 2.0–2.5 (WHO); 5.7 with 95% CI: 3.8–8.9 (CDC) |
| **Number of Reported Cases** | More than 8000 | 31.44 million through September 22, 2020 |
| **Number of Reported Deaths** | 774 | 967,197 through September 22, 2020 |
| **Mortality Rate**        | About 9%              | 3.1%                  |
| **Primary Mode of Transmission** | Infectious respiratory droplets dispersed from mucous membranes | YES                   |
| **Ability to Survive on Surfaces** | YES |                        |
| **Median Incubation Period** | 4–7 days             |                        |
| **Maximum Incubation Period** | 14 days              |                        |
| **Potential to cause severe respiratory infection** | YES |                        |
| **Potential to infect CNS and brain** | YES |                        |

Differences between SARS and COVID-19
SARS had a mortality rate of about 9%, which is 4–10 times higher than that of COVID-19. Unlike SARS-CoV-2, there were no reports of SARS-CoV transmission before symptoms appeared, and mild SARS-CoV infections were believed to be not transmissible (Table 1) (7) The basic reproduction number, R_0, defined as the average number of secondary infections produced by an infected person, is used to describe the transmission potential of infectious diseases. Using the World Health Organization (WHO) estimates, Petrosillo predicted that the R_0 for SARS is in the range of 1.7–1.9, whereas, for COVID-19, it was predicted to range between 2.0 and 2.5 (8) However, the Canters for Disease Control and Prevention (CDC) estimated that the R_0 for COVID-19 is much higher (5.7; 95% CI: 3.8–8.9) (9) A larger difference in the frequency of COVID-19 cases compared to SARS cases and its ability to spread rapidly across the globe indicates that the R_0 value for COVID-19 is most likely closer to the CDC estimate (Table 2).
Table 2. Similarities and Differences between SARS and COVID-19

|                        | SARS                  | COVID-19              |
|------------------------|-----------------------|-----------------------|
| Pre-Transmissibility   | NO                    | YES                   |
| Mild Case Transmissibility | NO                  | YES                   |
| Reproduction Number (R0)| 1.7-1.9 (WHO)        | 2.0-2.5 (WHO); 5.7 with 95% CI: 3.8-8.9 (CDC) |
| Number of Reported Cases | More than 8000       | 31.44 million through September 22, 2020 |
| Number of Reported Deaths | 774                 | 967,197 through September 22, 2020 |
| Mortality Rate         | About 9%              | 3.1%                  |

Purple-shaded items indicate similarity; yellow-shaded represent relative levels, and blue-shaded items are unique to COVID-19.

SARS-COV-2 TRANSMISSION

Transmission of SARS-CoV-2 from the Patients to the Host. Frequent sneezing and dry coughing exhibited by the COVID-19 patient generate viral plumes of thousands of droplets per cubic centimetre. Since SARS-CoV-2 infection is believed to be transmitted by aerosols and/or droplets, it is imperative to assess their particle characteristics, aerodynamic behaviour, and their propensity to bypass various physiological barriers to enter the host body (7).

![Diagram of Host receptor interaction with the SARS-CoV-2 spike protein and subsequent viral cell fusion with the host cell membrane.](image_url)

**Fig 2:** Host receptor interaction with the SARS-CoV-2 spike protein and subsequent viral cell fusion with the host cell membrane.
DIAGNOSTICS
Overview of SARS-CoV-2 detection
The first step in managing COVID-19 is the rapid and accurate detection of SARS-CoV-2 enabled by real-time reverse transcription–polymerase chain reaction (RT–PCR). RT–PCR detects SARS-CoV-2 nucleic acids present in nasopharyngeal fluids. Testing is used to prevent infectious spread between persons and communities that include asymptomatic infected persons, whose viral shedding can inadvertently spread the infection to the elderly and those with disease comorbidities. Accurate viral detection is a starting point to contain the COVID-19 pandemic10. Lapses affect public safety, enabling infection spread aided by false-negative test results. Improving test sensitivity and specificity remains an urgent need. Serological testing complements virus detection, indicating past infection, which could be harnessed for therapeutic gain. Antibodies are detected by enzyme-linked immunosorbent assay using a qualitative detection of IgG or IgM antibodies. Such tests determine an immune response against the viral spike (S) protein and may be helpful to assess protection against subsequent viral exposure and/or for contact tracing purposes. Thus, the importance of such tests cannot be overstated. This is also true for epidemiological evaluations and broad global therapeutic needs14. Future work includes the development of diagnostic tests to improve immunoassay sensitivity and specificity. Indeed, such testing will ultimately reveal viral protection as reinfections emerge. Inducing immunity against SARS-CoV-2 is the next frontier for COVID-19 control. To this end, our intent in this Review is to summarize the clinical disease presentation with a focus on how to best deploy nanomaterial-based and other diagnostic tests at individual, community and societal levels. The Review outlines current and future nanomaterial diagnostics for COVID-19. The intent is to facilitate the containment of the virus’s global spread (8).

Detection of SARS-CoV-2 viral shedding
In throat swabs and sputum, the viral shedding peaks at five to six days after symptom onset and ranges from 10^4 to 10^7 copies ml^-1. This reflects higher virus levels in the respiratory tract21. The viral RNA detection rate in nasal swabs of infected people has approached 100%. The positivity rates for blood, saliva and tears are 88, 78 and 16%, respectively. The self-collection of naso- or oropharyngeal swabs facilitates large-scale population field testing employing the chemiluminescence immunoassay and the enzyme-linked immunosorbent and lateral-flow immunochromatographic assays. The lateral-flow immunochromatographic assay uses gold nanoparticles (AuNPs) and a colorimetric label to provide a rapid platform for point-of-contact serological detection. Here, SARS-CoV2-specific antigen is conjugated with nanoparticles. By blood or saliva specimen loading, SARS-CoV-2 IgG and IgM can bind to the SARS-CoV-2 antigen and antibody, which is detected colorimetrically. The assay is completed in 20min with a ~90% accuracy24. To date, the minimum length of viral shedding is 7d after symptom onset, with viral infectivity observed within 24h. SARS-CoV-2 detection declines to undetectable levels, parallel the presence of serum neutralizing antibodies. Even among cases with concurrent high viral loads, the live virus could not be propagated in cell culture 8d after symptom onset. These studies warrant the use of quantitative viral RNA load and serological assays when deciding whether to discontinue infection control precautions (9).

RT–PCR
Current diagnostic tests for the SARS-CoV-2 pandemic use nucleic acid, antibody and protein-based detections, but viral nucleic acid detection by RT–PCR remains the gold standard. Nucleic acid tests have improved sensitivity and specificity for viral detection over the now available serological tests. The recognition of SARS-CoV-2 over common respiratory pathogens is contingent on RT–PCR serving as a sensitive, precise and specific viral detection. Despite the test’s accuracy, results have not yet enabled the containment of viral infection. In February 2020, the US Food and Drug Administration (FDA) permitted licensed laboratories to report in-house SARS-CoV-2 diagnostic tests. The procedure begins with the isolation and conversion of viral RNA to complementary DNA (cDNA). Next, the cDNA is amplified using Taq DNA polymerase. The RT–PCR test’s final overall workflow, which quantifies the viral load, is illustrated in Fig. 3. The total turnaround time can exceed 2d and runs the risk of reduced specificity through cross-contamination. The tests are commonly performed in hospital laboratories. Results from real-time RT–PCR using primers targeting different viral genome parts can be affected by viral RNA sequence variation. In addition, false-negative results may occur because of viral evolution16. Other limitations of RT–PCR tests include sample storage, low-quality nucleic acid purification, cost and wait times. Despite such limitations, the RT–PCR test remains the gold standard for SARS-CoV-2 diagnostics. For the alternative in situ hybridization and immunohistochemistry, collection of large amounts of sample is required and this can generate aerosols and
safety limitations. Immunohistochemistry is dependent on the choice and specificity of the antibody and sample quality. The most definitive method for the virus is high-throughput sequencing, but this approach is limited due to the cost, equipment and skillsets required. Isothermal amplification is a useful alternative to thermal cycling-based nucleic acid amplification. Simplified RT–PCR is now available to detect diverse regions of the SARS-CoV-2 genome. These detect the RNA-dependent S and RNA polymerase (RdRp)/helicase (Hel) proteins and the nucleocapsid (N) genes of SARS-CoV-2. The RdRp/Hel assays are highly sensitive means for viral detection. This combined with proper handling of large sample numbers by automated solutions and cobas 6800 systems provides fast and reliable test results.

Fig. 3: SARS-CoV-2 serological testing. Commonly used immune-based tests contain SARS-CoV-2-specific recombinant antigens immobilized onto nitrocellulose membranes. Mouse anti-human IgM and IgG antibodies conjugated with coloured latex beads are immobilized on conjugate pads. The test sample contacts the membrane within the test. The coloured antibodies form latex conjugate complexes with human antiviral antibodies. This complex immobilized on the membrane is captured by the SARS-CoV-2-specific recombinant antigen. If SARS-CoV-2-specific IgG/IgM are present in the sample, this leads to a coloured band, indicating a positive test result. The complex is captured on the membrane by goat anti-mouse antibody, forming a red control line. A built-in control line appears in the test window. The absence of a coloured band demonstrates a negative result. a–e, The workflow begins with patient serum added to the sample flow well (S) (a), saline buffer is added dropwise (b), and the sample incubated (c) until antibody–antigen recognition (d) and SARS-CoV-2 antibody detection (e). f, The rabbit antibody–gold shows in the control (C) well. g, A positive test (T) band indicates: : :: the presence of COVID-19 antibody and results without a positive C band are invalid. Notably, this assay depicts a post-immune response and may show negative results for individuals who have been recently infected. It may also detect virus in previously infected but asymptomatic persons.

SARS-CoV-2 diagnostics using nanomaterials
Nanomaterial-based technology provides feasible alternatives to RT–PCR for quick and precise viral detection. For example, magnetic nanoparticles can facilitate viral RNA extraction through coprecipitation, followed by polyamine ester functionalization via (3-aminopropyl) triethoxysilane, and can be used for up to 50,000 diagnostic tests. Quantum dots (QDs) could serve as ideal detection tools to study S protein–ACE2 binding dynamics and internalization due to their relatively small size, photostability and the ease of surface functionalization with biological molecules for Forster resonance energy transfer biosensing systems with various energy transfer partners, such as AuNPs that are characterized by absorption of electromagnetic
radiation in the visible region of the spectrum. A colorimetric assay was developed based on thiol-modified antisense oligonucleotides conjugated with AuNPs for detection of SARS-CoV-2 N-gene RNA. This is used for rapid diagnosis and can be performed within 10 min. The lower limit of detection is 0.18 ng/μl RNA particles. A recombinant S receptor binding domain conjugated to fluorescent QDs was created as an imaging probe for energy transfer quenching with ACE2-conjugated AuNPs. Upon binding of the S to the ACE2 receptor, fluorescence is quenched by the nearby AuNPs to enable monitoring of the binding events in the solution. QD probes can also facilitate cell-based assay identification and validation of inhibitors of the SARS-CoV-2 S protein and ACE2 receptor binding. The QDs are used as probes to investigate other viral receptors. This system can identify neutralizing antibodies and recombinant proteins for SARS-CoV-2 and other viruses with S-mediated cell recognition and entry. Biosensors have been developed for detecting influenza, the human immunodeficiency virus and other viral diseases. Initially marred by low sensitivity and specificity, limitations were overcome by plasmonic (gold and silver), metal oxide nanoparticle and field effect transistor (FET) bio- and graphene sensors. Graphene has wide application; it consists of hexagonal carbon structures arranged in a two-dimensional sheet. This gives it a large surface area, high electronic conductivity and high carrier mobility, and graphene biosensors are highly sensitive. When developing a graphene-based biosensor to detect SARS-CoV-2, coronavirus S antibody was immobilized on a graphene surface using 1-pyrenebutyric acid N-hydroxysuccinimide ester linkers. This graphene was used as a sensing material in a FET device to detect the S up to 1 fg/ml concentration. The optical property of AuNPs and silver nanoparticles conjugated to antibodies, when they are bound to the viral antigens or RNA, causes a detectable signal, which can be used to detect SARS-CoV-2. Toroidal plasmonic metasensors were developed that detect a femtomolar concentration of the viral S protein. They showed that monoclonal antibody conjugation on functionalized AuNPs could be detected up to 4.2 nM concentration (lower limit of detection). Transmission spectra of metasensors can shift the excitation with a polarized beam of light at terahertz frequency. Metasensors can be very useful in point of care (PoC) testing, where a rapid and sensitive assay is required. Recently, researchers have devised a single-step, optical S-protein-specific nanoplasmonic resonance sensor that requires minimal sample preparation and provides fast and direct virus detection. In such a system, highly specific antibodies to SARS-CoV-2 were immobilized on nanosensor chip surfaces to which intact coronavirus particles bind through S protein, leading to plasmon resonance or intensity changes that can be optically measured through a sensing system. For this assay, the lower limit of detection is 30 virus particles. The assay can be completed in 15 min. The assay can quantify virus below standard nasopharyngeal swab and saliva viral concentrations. On analysing the specificity of the sensor for binding SARS-CoV-2 in comparison to SARS-CoV, Middle East respiratory syndrome coronavirus (MERS-CoV) and vesicular stomatitis pseudoviruses, nanoplasmonic sensor chips demonstrated very high specificity (>1,000:1) in detecting the SARS-CoV-2. The nanoplasmonic sensor chips have the advantage of being low cost and scalable while maintaining uniformity and repeatability. The design of periodic nanostructures, without any external coupling optics, allows sensor chips to be integrated with a standard 96-microwell plate or microfluidic cuvettes. This allows standard microplate reader measurements. A low-cost, portable device controlled using a smartphone application can analyze SARS-CoV-2 in one step within 15 min with sensitive viral detection. Although the detection limit is 370, the virus can be quantified linearly from 0 to 107 virus particles per millilitre and it may find application in clinics, roadside screening sites and homes. AuNP-based sensors coupled with artificial intelligence can detect volatile organic compounds associated with SARS-CoV-2 in exhaled breaths. The assay is able to detect virus on the basis of the change in resistance of the nanomaterial biosensor layer. The methods can be optimized in future months by using other nanomaterials and larger cohort testing. A clinical diagnostic sensor was developed that combines a dual-functional plasmonic photothermal effect with localized surface plasmon resonance sensing transduction. Tests are done on two-dimensional gold nanoislands. The gold nanoislands contain complementary DNA receptors, which hybridize to SARS-CoV-2 nucleic acids. This system can be excited at two different wavelengths as it uses two different angles of incidence, one from a plasmonic photothermal biosensor and the other from localized surface plasmon resonance. It can detect RdRp-COVID, F1ab-COVID and envelope (E) genes from SARS-CoV-2. The dual-functional localized surface plasmon resonance biosensor has a lower detection limit of 0.22 pM and allows precise detection of selected SARS-CoV-2 sequences in a multigene mixture. The plasmonic sensing system can significantly reduce the rate of false-positive results. Similarly, others developed a plasmonic nanohole array used to transmit light for the label-free detection.
of the pathogen in biological media without sample preparation. It can quantitate intact virions by capturing them on group-specific antiviral immunoglobulins immobilized at the surface of the sensor. The intact virus binds to a suspended nanohole array grating that couples incident light to surface plasmons, causing a redshift in surface plasmonic resonance frequency. The assay could detect small (vesicular stomatitis virus and pseudo typed Ebola and large (vaccinia virus) enveloped viruses. The non-destructive nature of the assay allows for further analysis of progeny virions. Overall, the biosensors and other material-science-based detection techniques can enable rapid and portable diagnostic SARS-CoV-2 testing.

Fig. 4 | RT–PCR and LAMP assays for detection of SARS-CoV-2 infection. a, The RT–PCR assay. (i) A nasopharyngeal swab collects patient samples. (ii), (iii) RNA is extracted from fluids that contain SARS-CoV-2-infected cells and free viral particles. (iv) The recovered viral RNA is then reverse transcribed to cDNA and amplified for detection of viral nucleic acids. Conserved regions of the RdRp and E genes are the subgenomic viral segments amplified with a fluorogenic probe by qPCR. (v) Positive cases exceed the threshold of detection. b, The SARS-CoV-2 RT–LAMP assay: (i) amplification mixtures; (ii) RT–LAMP reaction; (iii) the products of SARS-CoV-2 RT–LAMP reactions. Although RT–PCR methods are used as the standard for detection of SARS-CoV-2 due to high sensitivity, limitations are present. As an alternative, isothermal amplification or LAMP was developed. When optimized for detection, the assay is as sensitive as standard PCR, detecting <10 viral copies per reaction. dNTP, deoxyribonucleoside 5′-triphosphate. c, SARS-CoV-2 saliva test. The illustration demonstrates SARS-CoV-2 infection in salivary glands and the released specific biomarkers that accumulate in the oral cavity. These are collected through a sample tube, tagged with a specific biomarker protein and run through lateral-flow rapid tests. Bulbar conjunctival injection in SARS-CoV-2 infected individuals is common.
Detection of SARS-CoV-2 antibodies

The synthesis of antibodies against SARS-CoV-2 is a primary immune response to infection. Neutralizing antibodies are found in up to 50% of infected individuals by day 7 and in all infected individuals by day 14. Serological studies are an alternative to RT-PCR for SARS-CoV-2 diagnostics. Combining real-time PCR and serological testing significantly increases positive viral detection rates. IgM levels increase during the first week after SARS-CoV-2 infection, peak after 2 weeks and then fall back to near-background levels in most individuals. IgG is detectable after 1 week and is maintained at a high level for a long period. In contrast, IgG becomes detectable after 1 week, remains elevated for an extended period, sometimes even more than 48d, and may serve to protect against reinfection. IgA responses appear between 4 and 10d after infection. Notably, a diagnostic predictor is the presence of serum IgA as well as IgG and IgM. The spectrum of SARS-CoV-2 antibodies is explained, in part, by divergent target antigens. Antibody titres can decrease 7d after infection. Recent studies have identified SARS-CoV2-specific antibodies in the saliva. Multiplex SARS-CoV-2 antibody immunoassays were investigated to determine differences between antibody levels in saliva and sera. Antibodies in saliva consistent with those in sera suggest parallel compartmental humoral immune responses. A parallel study developed rapid immunoassay using the BreviTest platform technology for measuring salivary IgA, which correlates with COVID-19 disease severity. Interestingly, low levels of IgA were seen in individuals with IgG without known exposure to the virus, and suggest that it may represent an indicator of herd immunity. SARS-CoV-2-specific antibody detection, especially that in saliva, may be useful for surveillance. Questions remain as to which antigens are the best candidates for serological testing. While the viral S is perhaps the strongest candidate, what remains unresolved is what part of the S should be developed. Alternatively, multiple isoforms of the S protein, such as those found in variant strains, may be used to ensure assay reproducibility. Time to results can vary from 13min (Abbott ID NOW) to 45min (Cepheid Xpert Xpress). Of the five antibody-based tests available, two are lateral-flow immunoassays (BioMedomics rapid test and Sure Screen rapid test cassette), one is a time-resolved fluorescence immunoassay (Gold site diagnostics kit) and two are colloidal gold immunoassays (Assay Genie rapid PoC kit and Viva Diag COVID-19 IgG–IgM based). Clinical studies will be needed to determine their clinical relevance. For N-based immunoassays, SARS-CoV-2 IgG (Abbott) shows a sensitivity of up to 100%. For S-based immunoassays, Liaison SARS-CoV-2 S1/S2 IgG and the combination S- and N-based platform COVID-19 VIRCLIA IgG MONOTEST demonstrated equivalent sensitivities. The plaque reduction neutralization test showed a sensitivity of 93.3%. To evaluate specificity, all of the tests except one, the enzyme-linked immunosorbent assay (IgG) (EUROIMMUN), produced at least one positive result for the negative SARS-CoV-2 antigen control. This probably represents large discrepancies between the testing platforms and the assay sensitivity relative to time. Although the plaque reduction neutralization test is the gold standard for immunoglobulin-based detection, the test has constraints, including a limited number of sample analyses, and requires a biosafety level 3 laboratory. The titres obtained from the assays correlate well with the plaque reduction neutralization test. Currently, antibody assays are applied principally for epidemiological testing.
Fig. 5: Nanomaterial-based SARS-CoV-2 detections. Representative illustration of nanomaterial-based biosensors for SARS-CoV-2 detection. Top: a FET-based biosensing device for detecting SARS-CoV-2. The sensor was produced using graphene sheets with specific antibodies against the SARS-CoV-2 S protein. Bottom: the dual-functional plasmonic photothermal biosensor and localized surface plasmon resonance biosensor on two-dimensional gold nanoislands functionalized with either cRNA receptors for detection of the selected SARS-CoV-2 sequences by fluorescence and FRET-based nucleic acid hybridization or with nucleic acid or antibody-functionalized nanomaterials for SARS-CoV-2 detection by colorimetric and antigen-binding assays. Ab, antibody; Ag, antigen; FRET, Forster resonance energy transfer; NPs, nanoparticles. Schematic ideas and technical methodological details were followed as represented in previously published reports.

SARS-CoV-2 antigens

A rapid diagnostic assay was also developed to detect the presence of viral antigens expressed by SARS-CoV-2 in samples from the respiratory tract of infected individuals. For this assay, antigen present in the sample binds to antibodies affixed to a paper strip enclosed in a plastic casing. This reaction generates a visually detectable signal within half an hour. The detected antigen(s) are expressed only if the virus is actively replicating; therefore, the tests can be used to identify acute or early infection. Also, a more common type of rapid diagnostic assay, which detects the presence of antibodies in the blood of infected individuals, has been marketed for COVID-19 by Abbott. Abbott’s test can detect the SARS-CoV-2 antibody on ARCHITECT i1000SR and i2000SR.
laboratory instruments, which can run ~100–200 tests per hour. Antibodies against SARS-CoV-2 are produced after one week of infection. The strength of any antibody response depends on age, nutritional status, disease severity, comorbid conditions and medications.

**Radiographic testing**

Although quantitative and qualitative tests of viral nucleic acid RT–PCR tests are the primary assay for SARS-CoV-2 detection, the sensitivities of these tests remain low for oropharyngeal (32%) and nasal (63%) swab samples. RT–PCR tests can often take up to more than a week due to a shortage in testing supplies or lack of technical skills. Therefore, suspected cases, either with or without RT–PCR results, require additional affirmations. Combinations of radiographic, molecular and antigen-based assays have been used alone or in combination to determine the optimal means to make a definitive diagnosis of SARS-CoV-2 infection. After the respiratory symptom presentation and nucleic acid viral detections, an initial evaluation of patients with COVID-19 commonly includes radiological examinations. Such examinations include a chest X-ray (CXR), CT or lung ultrasound (LUS). These, alone or together, can be also be used to stage SARS-CoV-2 infection. Often, a simple CXR is sufficient. However, a negative CXR alone cannot rule out lung involvement. While RT–PCR remains the gold standard for a virologic diagnosis, a CXR affords 69% sensitivity. However, imaging features contained in a standard CXR are often non-specific. When radiographic features of the disease are seen they reflect dense radiological patches on the left upper lobe and lower corners of the lung. With disease progression, more well-defined radiographic features are present and increase the veracity of a definitive COVID-19 diagnosis. However, while a CXR is the most useful test to affirm lung disease, it does not rule out alternative infections, especially in the context of presenting COVID-19 signs and symptoms, since it is not specific. Abnormalities such as pneumothorax, pulmonary oedema, pleural effusions, lung mass or lung collapse are alternatives. The value of the CXR is further supported by meta-analyses of patients with lower respiratory infections, including those treated in an intensive care unit. Serial chest X-rays can shorten symptom duration and reduce disease.

**Ultrasound**

Ultrasonography of the lungs is also used to assess patients with COVID-19. LUS does not appear to have specificity for identifying COVID-19 pneumonitis or pneumonia but is recommended for defining the area of infection. LUS may be beneficial in the early diagnosis of COVID-19 pneumonia as a cost-effective way to determine the localization of infection. The result of LUS is more sensitive than a CXR due to its excellent response to positive end-expiratory pressure (PEEP: pressure in the lungs above atmospheric pressure that exists at the end of expiration). LUS shows several features, such as lung consolidation in severe local disease. Similar to that found with CXR or chest CT, LUS in patients with COVID-19 infection found more prominent evidence of COVID-19 pathology in the posterior lower lung zones. In most cases, the infection progresses from the periphery to the centre of the lung tissues. Intensive care unit teams also use LUS findings of pulmonary oedema to therapeutically position the patients.

**TREATMENTS FOR COVID-19. Antiviral agents**

Currently, no specific anti-SARS-CoV-2 agent has been approved for treating COVID-19 patients. With only in vitro findings (for the SARS-CoV-2 and/or related viruses) and a lack of clinical experiences, several antiviral agents have been used under the “compassionate use” principle in the United States as they undergo testing in clinical trials.

**Lopinavir and ritonavir**

Lopinavir and ritonavir have been approved by the Food and Drug Administration (FDA) for treating human immunodeficiency virus (HIV) infection. The agents have also been used in the empirical treatment of SARS, and they are currently under study for the treatment of MERS because lopinavir is a promising agent that suppresses the protease activities of CoVs in vitro. In China, these two antiviral agents have been used to treat some COVID-19 patients in concomitant with alpha interferon. Han and co-workers reported a case study of a 47-year-old man with a 1-week history of cough, fever, and bosom frowsty. The patient had a history of stage two hypertension and type II diabetes. To resolve phlegm, relieve asthma and suppress virus replication, this patient was treated with 800 mg of lopinavir, 200 mg of ritonavir, 10 million IU of interferon alfa-2b recombinant, 400 mg of moxifloxacin hydrochloride, 60 mg of ambroxol hydrochloride, and 40 mg of methylprednisolone each day. The next day, the patient had low-grade intermittent fevers (36.0 °C to 37.2 °C). Moreover, many symptoms such as cough with production of phlegm/mucus, runny or stuffy nose, fatigue and vertigo were considerably alleviated. Due to the repeated negative testing results on days six and seven and in addition to the partial absorption of lung lesions, the patient was no longer infectious and discharged from hospital on day 10, suggesting that
the treatment strategy could be effective. A recent review found that the anti-CoV effect of lopinavir/ritonavir combination therapy mainly occurred with early treatment window and included the reduction of patient fatality and glucocorticoid administration. However, if the ideal window was missed, the treatment might have no significant effects on patient outcomes. Therefore, further studies that explore the clinical effects of the early use of lopinavir/ritonavir combination therapy for treating SARS-CoV-2 pneumonia are needed.

**Remdesivir**

Remdesivir, a monophosphoramidate prodrug of C-adenosine nucleoside analogue, can be incorporated into viral RNA chains and thereby initiating the premature termination of RNA replication. It displays a significant anti-CoV activity in vitro. Previous studies showed that remdesivir could inhibit the replication of SARS-CoV, MERS-CoV, and bat CoV strains in primary human airway epithelial cells and regulate cell entry through hACE2 receptor. Remdesivir acts during early-stage infection and dose dependently reduces RNA levels, which parallels a decrease in virus titers. Remdesivir displays an EC90 (90% effective concentration) value of 1.76 μM towards SARS-CoV-2 in Vero E6 cells, indicating that it is effective in non-human primates. Besides, it is noted that SARS-CoV-2 requires RdRp gene to replicate, which can be covalently bound to remdesivir and hence terminating chain elongation. Moreover, remdesivir also suppresses virus infection in human liver cancer Huh-7 cells, a cell line that is susceptible to SARS-CoV-2. Furthermore, remdesivir has been given on a compassionate use basis to COVID-19 patients, and the results indicated that a 10-day course of the antiviral drug (200 mg on day 1, followed by 100 mg daily for 9 days) might exhibit potential clinical benefit for these patients. Chloroquine Chloroquine is a potent broad-spectrum antiviral agent, which is commonly utilized as an autoimmune disease or anti-malarial agent. Chloroquine may stop a virus infection by elevating the endosomal pH necessary for virus-cell fusion and disrupting the terminal glycosylation of hACE2 receptor. Recent work has shown that chloroquine inhibits SARS-CoV-2 infection at entry and post-entry stages in a Vero E6 cell line. After oral administration, chloroquine is distributed throughout the body, especially in the lungs. Apart from its antiviral activity, chloroquine also exhibits immunomodulatory activities, which in turn lead to a synergistic enhancement of its antiviral effect in vivo. Chloroquine has been considered as a potential drug for the treatment and prevention of COVID-19 pneumonia. However, a recent study has found that chloroquine is ineffective for the treatment of COVID-19. Given the controversy about the effectiveness of this antiviral agent, it is important to determine whether chloroquine has potential applicability for SARS-CoV-2 treatment and prevention.

**Arbidol**

Arbidol is a broad-spectrum antiviral agent that blocks influenza A and B viruses by inhibiting virus-cell membrane fusion. Low-level evidence shows that arbidol taken alone or concomitantly with other antiviral drugs produces therapeutic benefits for COVID-19 pneumonia. In China, many randomized control trials are currently underway to assess the efficacy of arbidol on COVID-19 pneumonia. Performed a retrospective cohort trial on healthcare workers (n = 124) and family members (n = 66 members in 27 families) who have been exposed to a confirmed case of COVID-19. The authors found that arbidol could decrease the risk of SARS-CoV-2 infection in both hospital-care and family-care settings. Compared the efficacy of arbidol and lopinavir/ritonavir in 50 patients with laboratory-confirmed COVID-19, and the results demonstrated that COVID-19 patients treated with arbidol recovered more rapidly than those treated with lopinavir/ritonavir (P<0.01).

**CONCLUSION:**

The SARS-CoV-2 pandemic follows a troublesome trajectory. The health, humanitarian, social and economic policies adopted by countries can influence the speed and strength of the recovery. Currently, no medication is usually recommended to treat COVID-19, and no cure is accessible. The US FDA has authorized medicines previously approved for other disorders to be used now as recommended therapy for COVID-19. The earlier lack of accessibility for testing has hampered the infection control; however, testing of this novel virus is increasing quickly. Diagnostic testing for COVID-19 is vital in detection of the virus, understanding its epidemiology, case management and suppressing transmission. Universal operating procedures and harmonization of the available diagnostic assays are needed for faster screening approaches in the global fight against the pandemic. Similarly, academic scientists and biotechnologists are charged with the description of additional SARS-CoV-2 strains to improve the cluster-based specificity and sensitivity of antibody and antigen-based tests. Significantly, nanomaterial-based virus detection technology can help in the development of high-sensitivity, simple, scalable, rapid and cost-effective COVID-19 detection tests that supply on-demand
diagnostic capability effectively in the pandemic. This Review offers a road map for diagnostic strategies in the context of disease transmission and prevention. It is a basic science guide to better appreciate COVID-19 diagnostic complexities and to effect improved disease-combating strategies.

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