Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Biosensors and Bioelectronics 177 (2021) 112969

Contents lists available at ScienceDirect

Biosensors and Bioelectronics

journal homepage: http://www.elsevier.com/locate/bios

Biological characteristics and biomarkers of novel SARS-CoV-2 facilitated rapid development and implementation of diagnostic tools and surveillance measures

Gajanan Sampatrao Ghodake [a], Surendra Krushna Shinde [a], Avinash Ashok Kadam [b], Rijuta Ganesh Saratale [b], Ganesh Dattatraya Saratale [c], Asad Syed [d], Abdallah M. Elgorban [d], Najat Marraiki [d], Dae-Young Kim [a,*]

[a] Department of Biological and Environmental Science, Dongguk University-Seoul, Medical Center Ilsan, Goyang-si, 10326, Gyeonggi-do, South Korea
[b] Research Institute of Biotechnology and Medical Converged Science, Dongguk University-Seoul, Ilsandong-gu, Goyang-si, 10326, Gyeonggi-do, South Korea
[c] Department of Food Science and Biotechnology, Dongguk University-Seoul, 32 Dongguk-ro, Ilsandong-gu, Goyang-si, 10326, Gyeonggi-do, South Korea
[d] Department of Botany and Microbiology, College of Science, King Saud University, P.O. Box 2455 Riyadh, 11451, Saudi Arabia

ARTICLE INFO

Keywords:
SARS-CoV-2
RT-PCR
Diagnostic technologies
Point-of-care
Public surveillance
Mobile-health

ABSTRACT

Existing coronavirus named as a severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) has speeded its spread across the globe immediately after emergence in China, Wuhan region, at the end of the year 2019. Different techniques, including genome sequencing, structural feature classification by electron microscopy, and chest imaging using computed tomography, are primarily used to diagnose and screen SARS-CoV-2 suspected individuals. Determination of the viral structure, surface proteins, and genome sequence has provided a design blueprint for the diagnostic investigations of novel SARS-CoV-2 virus and rapidly emerging diagnostic technologies, vaccine trials, and cell-entry-inhibiting drugs. Here, we describe recent understandings on the spike glycoprotein (S protein), receptor-binding domain (RBD), and angiotensin-converting enzyme 2 (ACE2) and their receptor complex. This report also aims to review recently established diagnostic technologies and developments in surveillance measures for SARS-CoV-2 as well as the characteristics and performance of emerging techniques. Smartphone apps for contact tracing can help nations to conduct surveillance measures before a vaccine and effective medicines become available. We also describe promising point-of-care (POC) diagnostic technologies that are under consideration by researchers for advancement beyond the proof-of-concept stage. Developing novel diagnostic techniques needs to be facilitated to establish automatic systems, without any personal involvement or arrangement to curb an existing SARS-CoV-2 epidemic crisis, and could also be appropriate for avoiding the emergence of a future epidemic crisis.

1. Introduction

A severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) was first appeared in the China Hubei Province, Wuhan at the end of December 2019. Considerable number of sick patients with severe and moderate symptoms including fever, shortness of breath, and coughing were rushed for admission to the nearby hospitals. These patients were underwent computed tomography (CT) scans and the results revealed opacities in their lungs (profuse, dense, and confluent types), which were differed from that of the CT scan images of the healthy human lungs (Ai et al., 2020; Zhou et al., 2020c). Ahead of the development, existing nucleic acid-based diagnostic kits, CT scans, and symptoms were collectively used in the initial diagnosis of SARS-CoV-2 infections. Later, well-established nucleic acid-based test kits were made available for most of the known viral panels and performed with a straight multiplex approach using a well-known technique called real-time polymerase chain reaction (RT-PCR), but, the results were found to be negative, indicated that the contagion of the infection was novel and thus, the origin of virus was unknown (Park et al., 2020; Zhang et al., 2020d). In the first week of January 2020, bronchoalveolar lavage (BAL) fluid samples of different patients were examined and mysterious virus with great similarity to the viral genome of the betacoronavirus-B family

* Corresponding author.
E-mail address: sbpkim@dongguk.edu (D.-Y. Kim).

https://doi.org/10.1016/j.bios.2021.112969
Received 10 September 2020; Received in revised form 30 December 2020; Accepted 2 January 2021
Available online 4 January 2021

was identified.

Thus, emergence of an extremely infectious novel coronavirus and its immediate reach worldwide has led to an unexpected emergency in public-health and global economy. Phylogenetic investigations of novel SARS-CoV-2 suggested that it belongs to the beta-coronavirus genus (Zheng 2020), which is of SARS-CoV and middle east respiratory syndrome (MERS)-CoV, and some of the other coronaviruses related to bats and pangolins (SARS-related coronaviruses, SARSr-CoVs), as well as some other coronaviruses known to infect a diverse range of mammalian species including humans (Pal et al., 2020; Wu et al., 2020a). The family coronaviridae with the closest similarity of SARS-CoV-2 seems that the Bat RaTG13 coronavirus has more than 93.0% similarity in the sequence of spike protein (S gene). However, many other types of SARS-CoV were also found to be very related from SARS-CoV-2, but, sequence similarity was only about 80% (Zhou et al., 2020b). Recent investigations revealed that this novel SARS-CoV-2 has ~50%, ~80%, and ~96% similarity index with the genome sequence for MERS-CoV, SARS-CoV, and the coronavirus bat-RaTG13, respectively (Lu et al., 2020a; Udugama et al., 2020; Zhou et al., 2020b).

As of December 30, 2020, the SARS-CoV-2 pandemic has spread globally to more than 220 countries and territories, confirmed infections about more than 82 million and caused deaths globally about 1, 80,000 in number (https://www.worldometers.info/coronavirus/). The number of reported SARS-CoV-2 infections are undoubtedly underestimated since there are millions of asymptomatic or mild cases those gone undetected and unreported (Long et al., 2020c; Tan et al., 2020a). Most of the asymptomatic individuals in Yokohama, Japan located on the Diamond Princess Cruise Ship were found to be an infectious, suggesting that the existence of a larger percentage of infected population and eventually recovered than that of confirmed and reported cases (Mizumoto et al., 2020).

The development of vaccines and therapeutics is ongoing in a rapid pace, but the United States Food and Drug Administration (FDA) is proactively involved in accepting submission and providing approvals for many of the vaccines and drugs those are in research and development (R and D) to handle existing SARS-CoV-2 pandemic crisis (Liu et al., 2020a). Early diagnosis and robust contact tracing as performed by South Korean administration, revealed as an important approach in suppressing the spread of SARS-CoV-2 infections. This initiative facilitated rapid implementation of administrative control measures that curbed further spread of SARS-CoV-2 infections through individual case documentation, monitored isolation, and be informed about case to case travel history to avoid direct or indirect contact with a confirmed patient (Ryu et al., 2020). Both developed and developing nations needed to perform surveillance measures in any legitimate manner possible to manage the existing outbreak and to combat the disease spread with a prompt public health prevention measures and intervention agencies in the remote areas with involvement of experts or any trained professionals.

In the COVID-19 era, the implementation of smartphone surveillance and mobile health (mHealth) technologies to assist recent developments in plug-and-play diagnostics will contribute significantly because of their anticipated affordability for on-site early diagnosis, as well as dampening the severity of infectious pandemic outbreak. The recently established diagnostic tools for SARS-CoV-2 and their testing workflow, strategies to curb infections are illustrated in Fig. 1. This review report devotedly summarized all aspects of recently reported biological characteristics, diagnostic technologies, and clinical markers, emerging diagnostic methods, and surveillance strategies for the existing SARS-CoV-2 epidemic. These topics are of great interest to curb rapidly spreading SARS-CoV-2; as a result, this review article is intended to summarize recent findings that will be useful as guiding principles and strategies to address the existing pandemic crisis caused by the SARS-CoV-2 coronavirus.

2. Biological characteristics of the SARS-CoV-2

Main structural features of the SARS-CoV-2 was appeared to be identical with other reported coronaviruses. The transmission electron microscopy (TEM) was successfully used to image the SARS-CoV-2 viral structures (Fig. 2). TEM imaging showed that the size of novel virus is ranging from the 60–140 nm in diameter and the RNA genetic material is packed inside the envelope and surface is decorated with the spike glycoproteins (Zhu et al., 2020a). This virus has a genome comprised of single-stranded positive-sense RNAs and nucleotides with a length of about ~30,000 (Mousavizadeh and Ghasemi 2020; Wu et al., 2020a; Zhou et al., 2020b). The viral genome encodes different proteins about 27, comprising an RNA-dependent RNA-polymerase (RdRP), in addition to the four other main proteins responsible for structural integrity. RdRP is well known to interact particularly with non-structural proteins and plays a role in sustaining the fidelity of the genomic materials (Denison et al., 2011; Jain et al., 2020; Wu et al., 2020a). Genome sequence analysis revealed that the RdRP gene of SARS-CoV-2 seems greatly identical with bat coronavirus RaTG13, with approximately about 96% of similarity (Zhou et al., 2020b). The four different structural proteins of SARS-CoV-2 are nucleocapsid (N), envelope (E), matrix (M), and surface spike glycoprotein (S) (Fig. 2). The S gene encoded in coronaviruses attributes to the S protein of the receptor-binding domain (RBD),

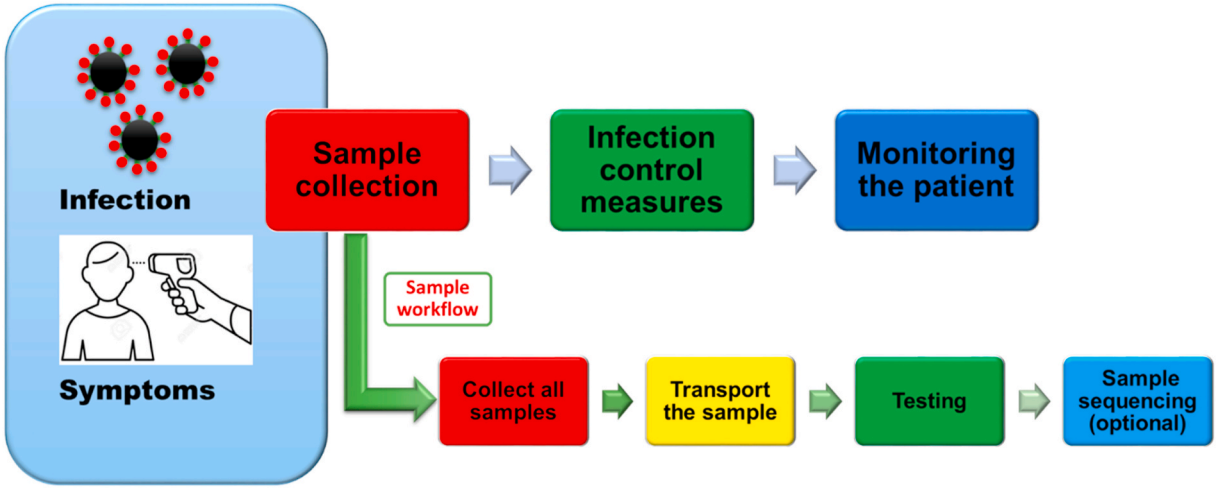

Fig. 1. Illustration of the SARS-CoV-2 diagnostic development and testing samples workflow during the course of the outbreak. Healthcare providers should provide triage facilities to manage patients appropriately. Samples can either be tested on-site or transported to a central facility for nucleic acid testing.

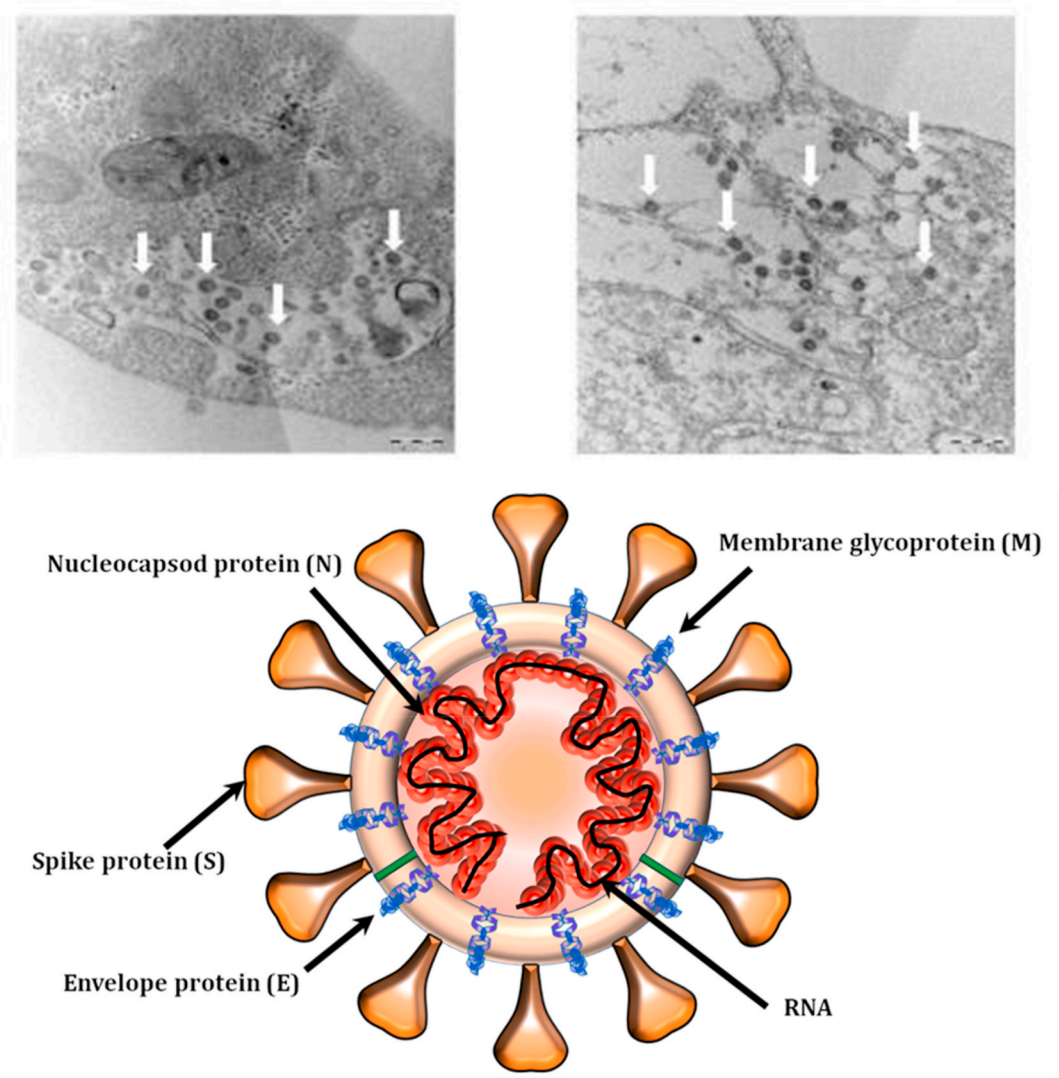

Fig. 2. The phenotype of novel SARS-CoV-2 virus, thin section of transmission electron microscope micrograph of Vero cells after infection. Reprinted with the permission of reference (Kim et al., 2020). Structural features of the SARS-CoV-2 virus are illustrated to represent different structural proteins intracellular genetic materials.

which assists the SARS-CoV-2 virus while bonding with the host cell receptor (Wrapp et al., 2020). S protein thus helps to mediate interaction with an angiotensin-converting enzyme 2 (ACE2) a host receptor, membrane fusion, viral entry, and also determines host tropism and basic reproduction number (Lu et al., 2020b). The S gene is quite diverse; about <75% of the nucleotide sequence is similar in previously reported SARS-coronaviruses and the rest of the sequence is variable (Cui et al., 2019). The remaining three structural proteins are generally known for conserved sequence in comparison to the S protein and are essential to undertake overall functions in coronaviruses (Wu et al., 2020a). The S gene also supports the encasing of the RNA and protein assembly, thus, formation of the envelope, the budding process, and further pathogenesis (Neuman et al., 2011; Schoeman and Fielding 2019).

2.1. Structure and function of receptor-binding domain

This report summarizes the recently reported genomic data and presents different scenarios through which SARS-CoV-2 became compatible with the human cell receptors. Comparative analysis of beta- and alpha-coronaviruses have revealed distinct genomic features for novel SARS-CoV-2. Initially it was identified through biochemical experiments (Letko et al., 2020; Wrapp et al., 2020) and structural analysis (Krafcikova et al., 2020; Shereen et al., 2020; Wan et al., 2020); thus, it suggested to be the first mutated feature and is appropriate for interaction with the ACE2 human cells receptors. And the second notable feature revealed on the basis of the S proteins found in the SARS-CoV-2 virus, which consisting a functional-polybasic “furin” cleavage site.

S protein is part of RBD sequence and is the greatly diverse region of the coronavirus genome and defines most of the virus features, in addition to the susceptibility, host range and cellular tropism (Lan et al., 2020; Wu et al., 2020b; Zhou et al., 2020b). Six amino acids in the RBD sequence are confirmed for their critical interaction with the ACE2 human host receptors and thus, adapts to the new host range of beta-coronaviruses (Wan et al., 2020; Yi et al., 2020). As reported previously, sequences are Y4911, N479, L472, D480, Y442, and T487, which attributes to Y505, Q493, F486, S494, L455, and N501, respectively for the SARS-CoV-2 (Wan et al., 2020). Five out of six amino acid residues differs between SARS-CoV and SARS-CoV-2 (Tang et al., 2020). Based on recent revelations, the SARS-CoV-2 has a unique RBD unit with great affinity to the ACE2 receptor of the human cells, ferrets, cats, and some other mammalian species are those with high receptor homology (Andersen et al., 2020; Damas et al., 2020; Shi et al., 2020). Thus,

scientists are now seeking a new model well suited for R and D activities, vaccines and treatments for COVID-19, and some data have been established on the ferrets those can effectively replicates the novel strain SARS-CoV-2 (Richard et al., 2020).

Computational analysis of S protein present in RBD of SARS-CoV-2 revealed that it has a potent ability to bind to ACE2 receptor in human lung cells with high affinity (Spinello et al., 2020). This interaction is now thought that it must quite unique from the RBD of the SARS-CoV that was also appropriate for ACE2 receptor binding (Sheahan et al., 2008; Wan et al., 2020). Therefore, the high binding affinity evolved in the SARS-CoV-2 at S protein of RBD and ACE2 human host receptor, in all likelihood, must be the consequence of natural selection that finally led to the explosion of the pandemic worldwide. This is also a strong indication that the SARS-CoV-2 must not be the creation of artificial mutation or manipulation through genetic engineering experiments or purposeful manipulation in a laboratory (Schröder 2020).

Another most prominent characteristic of SARS-CoV-2 is that contains polybasic (furin) cleavage site located at the S1 and S2 junction, which are the two main subunits of the S glycoprotein. This facilitates functional polybasic (furin) cleavage site and some of the other protease enzymes to perform determining function in the viral susceptibility in addition to the host-range of the coronavirus. In addition, a proline residue was also reported in this site of SARS-CoV-2 and responsible for the forming proline mediated turns that also predictable to result in auxiliary O-linked glycans to S673, S686, and T678, those are at the edge of this site and which also found exclusively in SARS-CoV-2. These sites were found identical to those for SARS-CoV-2, but not been reflected as a completely identical to betacoronaviruses of lineage B, though other human beta-coronaviruses, as well as human coronavirus HKU1 (lineage A), known to have such cleavage sites and are reported to consist of O-linked glycans (Chan et al., 2008). As genetic variations found in the S proteins, it was possible that the SARS-CoV-2 is a novel coronavirus with a fraction or full of polybasic (furin) cleavage site and also needs to be investigated for the some of the other coronaviruses.

Thus, the functional significance of polybasic (furin) cleavage site of SARS-CoV-2 got investigated immediately after the emergence, and it was most important achievement demonstrated their novelty and biological characteristics including, how this site alters the pathogenesis and transmissibility of the disease in a wide host range. Experimental evidences suggested that the SARS-CoV has its own unique mechanism of functioning through this site at S1 and S2 junction, which facilitates cell fusion, without disrupting the entry of viruses through the host cell membrane (Belouzard et al., 2009; Follis et al., 2006). Furthermore, an efficient polybasic (furin) cleavage site reported for S protein of MERS-CoV also known to makes vulnerable to MERS-like coronaviruses in camels, bats and some other host-ranges in addition to human host (Menachery et al., 2020). On the contrary, avian influenza viruses was known for rapid transmission and replication in chicken populations, where they attach to polybasic (furin) cleavage sites at the hemagglutinin (HA) proteins (Sonnberg et al., 2013), which functions similar to the S protein of SARS-CoV.

The function of O-linked glycans is yet to be fully revealed; it has been speculated to form a “mucin-like domain” that protects epitope or key amino acid residue sequence in the S proteins of SARS-CoV-2 (Bagdonaite and Wandall 2018), similar to those of several other viruses that function “mucin-like domains” as O-linked glycans shield, thus, intricate the host immune system evasion. Although O-linked glycans must be robust in SARS-CoV-2, to clarify further experiments are required in the direction of revealing the role of such sites. Furthermore, manipulation or possibility of creating of SARS-CoV-2 like virus in laboratory settings either by cell culture or animal host ranges requires a prior sequestration of ancestor viral strains those are with an identical genomic makeup, which is yet to be certainly revealed.

2.2. Receptor-binding domain–angiotensin converting enzyme 2 complex

SARS-CoV-2 binds with ACE2 human host receptors and enables entry into host cells. ACE2 receptors are present in almost all human organs as well as in the wide variety of cells including smooth muscle cells and endothelial cells of the lungs, stomach cells, colon, small intestine cells, liver cells, kidney cells, and also brain (Shereen et al., 2020; Zhang et al., 2020a). Recent findings revealed key structural differences and the intrinsic sequence of the RBDs of both of the SARS-CoV and SARS-CoV-2 (Majumder et al., 2020). The precise understanding of the interaction of the SARS-CoV-2 virus with ACE2 human cells receptors is now revealed after detailed structural analysis RBD–ACE2 complex using X-ray crystallographic examinations (Lan et al., 2020). The molecular and structural analysis improved our scientific understanding of the molecular interactions and susceptibility to the host cells (Chen et al., 2020b). Such information provided precise targets to facilitate the preparation of SARS-CoV-2 neutralizing antibodies and development vaccines, which are required with a great urgency to fight the existing global crisis emerged after SARS-CoV-2 spread (Dong et al., 2020; Pinto et al., 2020).

Coronaviruses use their homo-trimeric spike glycoprotein (each spike monomer consists of an S1 and S2 subunit) on the virus envelope to bind with the ACE2 human host cell receptors. Thus, after binding with the ACE2 receptor a key initial step enables SARS-CoV to enter in host cells. The second event is the initiation of a cascade of molecular events to eventually lead to a cell membrane fusion between host cell membranes and viral particles, thus finally allows to host cell entry. The cryo-electron microscopic examinations performed for SARS-CoV S proteins and their interface between ACE2 receptors of host cells have revealed that such binding encourages the dissociation of the S1 subunit from the ACE2 receptor. However, the S2 subunit induces a transition from a meta-stable pre-fusion form to an extra-stable post-fusion form and both events are vital for the viral and host cell membrane fusion (Kirchdoerfer et al., 2018; Song et al., 2018). Recent studies on *in-vitro* binding demonstrated that the SARS-CoV-2 RBD predominantly binds to human ACE2 cell receptors owing to their high affinity even when there at a lower nano-molar concentration range of viral dose, signifying that the SARS-CoV-2 RBD is a main functional domain inside the S1 subunit and make highly susceptible to bind SARS-CoV-2 virus with the host cells through ACE2 host cell receptors (Letko et al., 2020; Walls et al., 2020).

The notable and main feature of the SARS-CoV-2 RBD–ACE2 interface, the complex is made of hydrophilic interactions exerted from amino acid residues. There were about two salt-bridges and thirteen different hydrogen bonds responsible to play a key role in the formation of the RBD–ACE2 interface of the SARS-CoV (Hou et al., 2010), similar to the three salt-bridges and thirteen hydrogen bonds involves in the formations of RBD–ACE2 interface of the SARS-CoV-2 (Shang et al., 2020b). The second identical feature seems to be the presence of various tyrosine residues which helps in the formation of hydrogen-bonding interfaces within residues that contain polar hydroxyl groups (Robson 2020a), role of such tyrosine residues also been described in other recent publications (Lan et al., 2020; Robson 2020b). The third identical feature is the chain of Asn-90-linked glycans in the ACE2 receptor which binds with the RBD of SARS-CoV. Similarly, the chain of Asn-90-linked lysosomal enzyme called N-acetyl-beta-D-glucosaminidase (NAG), –NAG–β-D-mannose may cooperate with the RBD of SARS-CoV-2 thus, play significant roles in forming RBD or SARS-CoV-2 and ACE2 complex (Lan et al., 2020; Li et al. 2005a, 2005b; Reguera et al., 2014).

Furthermore, such identical features reported in recent and previous reports demonstrates that the RBD of SARS-CoV and RBD of SARS-CoV-2 with their respective ACE2 interfaces have considerable similarities in terms of binding surface area, the number of amino acid residues involved in complex-formation with their respective hydrophilic interactions (Lan et al., 2020). However, a minor difference was reported in terms of ACE2 interactions at both outsides and inside the region of

the receptor-binding motifs (Lan et al., 2020; Wrapp et al., 2020). Such identical features strongly reveals that the evolution of novel SARS-CoV-2 either is from the SARS-CoV virus or independently from the intrinsic RBD structure, with minor improvement in the binding affinity for ACE2 human receptor (Andersen et al., 2020). However, SARS-CoV-2 does not form clusters similar to the reported for beta-coronavirus genus including SARSr-CoV and SARS-CoV (Rastogi et al., 2020).

SARS-CoV-2 has 89.8% homology with SARS-CoV in terms of spike S1 and S2 proteins subunits, which are involved in facilitating the fusion process and host cell entry. Both subunits directly employ host human ACE2 receptors after infection (Zhou et al., 2020b). Therefore, high structural similarity of RBD of SARS-CoV-2 with RBD of SARS-CoV suggests that they have high binding affinities to ACE2 are identical in nature owing to the equilibrium dissociation constant (KD) for ACE2 with RBD of SARS-CoV-2 and ACE2 with RBD of SARS-CoV (Tian et al., 2020; Walls et al., 2020). However, such findings were slightly different from recent reports suggesting SARS-CoV-2 have 20-fold higher binding affinities for ACE2 with spike trimer having KD about 14.7 nM, in comparison to that of ACE2 with RBD of SARS-CoV, with KD about 325 nM (Wrapp et al., 2020). Therefore, the binding affinity alone cannot be used to explain 10-20-fold higher transmissibility for SARS-CoV-2, which is considered liable for the emerging existing global crisis (Wrapp et al., 2020; Zheng 2020).

Other important feature of having a unique (furin) cleavage site (-RRAR-) within junction of the S1 and S2 subunit of S protein in the SARS-CoV-2 and also have a significant role while enabling the higher rate of transmission from infected human to healthy humans (Okba et al., 2020; Xia et al., 2020). Thus, the binding affinity of ACE2 with RBD of SARS-CoV-2 at the S1 subunit is ranging from 10 to 20 fold greater than that with the RBD of SARS-CoV at the S1 subunit (Xia et al., 2020). This increased binding affinity must be triggered a global pandemic crisis owing to higher transmissibility and infectivity for recently evolved SARS-CoV-2 virus as compared to those of SARS-CoV (El Zowalaty and Järhult 2020a, b; Ferretti et al., 2020; Yang et al., 2020d). The potential and most debated route for the global spread of SARS-CoV-2 also suggested that of discharge of the virus particles from the infected patient into the surrounding environment via diverse means, which is now at the peak of rigorous argumentum in an excuse of rapid transmission of SARS-CoV-2. However, this hypothesis warrants further investigations to illustrate the diverse functional routes, structural features and pathogenesis of prevailing SARS-CoV-2 virus.

2.3. Antigenicity of the SARS-CoV-2 spike protein

There is a shortage of specific therapy because of the rapid spread of SARS-CoV-2, huge efforts have dedicated to develop antiviral drugs, vaccines, and neutralizing antibodies (Lu 2020). Vaccines helps to prevent the spread of disease, it is largely reliant on instigates memory for neutralizing antibodies against specific epitopes present on the antigen. Among the main structural proteins, the S glycoprotein consists of RBD and helps to mediate the entering of coronavirus to host cells, which makes S protein a main antigenic target for vaccine and neutralizing antibodies (Tian et al., 2020).

A recent study that, even though S protein of SARS-CoV-2 exhibits high about 75.5% homology towards the S protein of SARS-CoV, novel SARS-CoV-2 epitopes attributes to 85.3% of almost all of the antibody epitopes, the RBD antibody epitopes about 85.7%, and in SARS-CoV-2 high-score antibody epitopes about 90.9%, suggesting noteworthy variations in the antigenicity (Zheng and Song 2020). This report further reveals that the antigenicity of S protein of SARS-CoV-2 is extraordinarily evolved and dominated with their novel antibody epitopes, which may provide potential leads to drive the R and D activities during the development of vaccines. In addition to the RBD, which was identified to the spectacle as the main target for generating responses of neutralizing antibody in human hosts (Robbiani et al., 2020), recently N-terminal domain (NTD) was also evidenced as a new vulnerable site in S protein of SARS-CoV-2 for instigating another type of neutralizing antibody and thus it could similarly function as a protein-based recombinant vaccine (Brouwer (2020); Chi et al., 2020; Liu (2020c)). As predictable, neutralizing antibodies specific for NTD can target against the S protein in both open and closed conformations (Chi et al., 2020). Furthermore, the apparent reports on the HR1 region and fusion peptide of SARS-CoV-2 structure accessible in literature could help to focus on S ectodomain trimer as well as its high conservation sequence among SARS-CoVs coronaviruses (Tortorici and Veesler 2019). This approach suggests that the possibility of making a good choice of immunogen candidates while designing epitope-specific vaccines and neutralizing antibodies aimed at targeting broad range of SARS-CoV (Pallesen et al., 2017). The design of vaccines specific to the epitope has recently proven as a successful strategy in producing neutralizing antibodies targeting glycoprotein of respiratory syncytial viral fusion, but, such neutralizing antibodies need to be isolated carefully from infected patients (Correia et al., 2014).

Unlike other-type full-sequence of SARS-CoV-2 S protein, the monomeric fragment has no ability to induce any infection-mediated antibodies or unnecessary immune or hyper-inflammatory responses (Quinlan et al., 2020; Yang et al., 2020a), all of such events could be possibly avoided to improve immunogenicity by the structure-based design of immunogen (Anasir and Poh 2019; Kwong 2017). On the other hand, full-length wide-type or soluble ectodomain form of the S protein for SARS-CoV-2 can ably trigger a strong immune response (Grifoni et al., 2020b), which was also confirmed to play a key role in controlling the severity of diseases caused by SARS-CoVs (Liu et al., 2017; Yong et al., 2019), including SARS-CoV-2 (Oja et al., 2020), and possibly also act as an imperative factor in determining the effectiveness vaccines designed against SARS-CoV-2 (Ahmed et al., 2020; Grifoni et al., 2020b).

The structural investigation of SARS-CoV-2 viral structures by cryo-electron microscope revealed the S protein trimer in recent studies (Walls et al., 2020; Yan et al., 2020). Furthermore, the structure of S protein was also examined by modeling of RBD, mainly to study receptor-binding motif (RBM), which directly involves in interacting with ACE2 (Wrapp et al., 2020). Despite the apparent cross-reactivity of SARS-CoV-2, deactivating performance in plasma/serum recovered from the patients after infection (Hoffmann et al., 2020; Tian et al., 2020), isolated monoclonal-antibodies seems to be unable in deactivating SARS-CoV-2 effectively (Wrapp et al., 2020). However, a detailed modeling analysis on the binding between the ACE2 host receptor and RBD of SARS-CoV-2 has yet to be accomplished, it may reveal some more amino acid residues that may be involved in such interactions. The actual amino acid residues that facilitate the interactions yet remained unclear, as suggested in a recent report (Wan et al., 2020). The strategies for immunogen design are explicitly described in a recently published review report on the S glycoprotein of SARS-CoV-2 covering its function, structure, biosynthesis, immunogenicity as well as antigenicity in anticipation to contribute the development of effective and safe vaccine against SARS-CoV-2 emergency (Duan et al., 2020). This report covers recent understandings of many other biological characteristics of SARS-CoV-2 that have certainly empowered investigators to rapidly develop reliable diagnostics kits, some other technologies to perform point-of-care detection, and smartphone surveillance.

3. Diagnostic technologies for SARS-CoV-2

The symptoms experienced by SARS-CoV-2 patients are common pneumonia-like diseases and not applicable to use for an accurate diagnosis. It was reported that about 44% of 1099 surveyed SARS-CoV-2 patients from China on the onset of spread showed mild fever like symptoms before admission to the hospital, but, more than 80% of the confirmed patients then showed fever-like symptoms after the admission (Guan et al., 2020). Further examination of those admitted patients

revealed that about 68% of patients were developed series of symptoms of respiratory infections in addition to cough, sputum formation (34%), tiredness (38%), and about 19% of breathing problems. However, many such symptoms are also associated with pneumonia-like diseases. Before the establishment of nucleic acid-based testing kits using real-time reverse transcription polymerase chain reaction (RT-PCR) and mostly CT scans have been employed as the primary diagnostics for screening and diagnosing SARS-CoV-2 infections.

3.1. Nucleic acid-based diagnostics for SARS-CoV-2

Numerous RT-PCR kits were promptly developed and used to diagnose the SARS-CoV-2 patients. The RT-PCR method typically involves the reverse transcription process for the extracted RNA materials converts into complementary DNA strands (c-DNA), and then amplification of those derived cDNA copies (Freeman et al., 1999; Kageyama et al., 2003). The RT-PCR method depends on the capability of amplifying a small concentration of viral nucleic acid materials collected in test vials and it is also known as a gold standard protocol in the confirming SARS-CoV-2 infections (Morales-Narváez and Dincer 2020). The RT-PCR process begins with the transformation of viral RNA into c-DNA through a reaction with reverse transcriptase (RNA-dependent DNA polymerase). At present, RT-PCR examinations developed for the diagnosis of SARS-CoV-2 patients requires swab test samples that are typically collected from the upper respiratory tracts. The RT-PCR assays are developed by Rutgers Clinical Genomics Laboratory also uses saliva samples (Carter et al., 2020). This method is much rapid and less hurting than that of other test sample collection routes. It is also possible to lower the exposure risks for healthcare providers and also allows performing large-scale testing capacity (Xu et al., 2020).

The RT-PCR process is generally divided into two main phases; the first consists of primer design and sequence alignment, while the second involves assay optimization and testing. It is necessary to align and analyze different types of coronavirus-related genomes before designing a set of probes and primers (Carter et al., 2020). Three main regions that are responsible for conserved genomic sequences from SARS-related viruses have been discovered: (1) the nucleocapsid-protein (N gene), (2) the envelope-protein (E gene), and (3) RNA-dependent RNA-polymerase (RdRP gene). Both RdRP and E gene showed higher analytical sensitivity for identification of SARS-CoV-2, with a low limit of detection (3.5 and 4 copies), respectively/reaction cycle; however, the N-gene showed lower analytical sensitivity about 8.5 copies/reaction cycle) (Udugama et al., 2020). The most important advantage of this assay is that it can be designed for two different target systems, serving either as a primer that can be universally used to detect different types of coronavirus including existing SARS-CoV-2 (Mollaei et al., 2020), or as the primer that can identify SARS-CoV-2 with high specificity (van Kasteren et al., 2020).

Once selecting designs for the probes and primers, the following step involves is the optimization of the assay condition (reagent concentration, temperature, incubation times, etc.) and finally repeated analysis by RT-PCR. The one-step and two-step assay procedures can be performed with RT-PCR, particularly in a one-step assay RT-PCR amplification operates normally, where reverse transcription step is combines, thus possibly providing better reproducibility in addition to ensuing preparation of rapid test reports (Dharavath et al., 2020). However, the challenge with this approach lies in optimization the amplification steps and reverse transcription, which occurs concurrently, possibly leads towards generating lower target amplicon. However, in two-step procedure, the assay reaction gets completed sequentially in separate test tubes (Wong and Medrano 2005), thus, this approach have better sensitivity and accuracy than those of a one-step assay procedure. However, the two-step assay also requires the series of optimization protocols for some of the additional parameters and also to reduce time-consumption (Carter et al., 2020; Wong and Medrano 2005). Additionally, control samples are prerequisites to be performed simultaneously to make sure the reliability and accuracy of the assay and to address any of the trial errors prevailing. Several different RT-PCR based testing kits and post-infection antibody diagnosis kits are promptly industrialized and being predominantly employed for accurate diagnosis of SARS-CoV-2 infections at a global scale and some of such technologies are still under validation and approval by the international authorities (Nuccetelli et al., 2020; Vandenberg et al., 2020).

Smyrlaki et al. has performed a single-reaction RT-PCR using nucleocapsid 1 primer-probe to record its cycle threshold (CT) values designed for the serial dilution of the media (Smyrlaki et al., 2020) and reported inhibitory effects in all tested three media samples and, notably, marked variations among the media samples. Long et al. in their recent report also compared RT–PCR method for its CT values in their examination of nasopharyngeal swab samples received from firstly positive 37 asymptomatic patients and 37 symptomatic individuals (Long et al., 2020b). That report revealed that the initial CT values for 37 asymptomatic patients and 37 symptomatic individuals seemed to be similar, the short-lived duration for viral shedding was observed about 6 days, whereas the long-lasting viral shedding duration was supposedly appeared to be about 45 days. It is important to note the observation on the asymptomatic group that had shown significantly longer viral shedding duration than those of the symptomatic group, but, quantifiable virus shedding cannot be equated with the viral infectivity, thus, detailed further evaluation is required to determine whether SARS-CoV-2 viral load can be correlated with other respiratory viruses suitable to grow in cultures (Atkinson and Petersen 2020). In one of the prominent reports this was performed, using real-time PCR with reverse transcription examination showed the lowest CT values for swab samples collected from upper respiratory tracts that often possible to detect in the first week of SARS-CoV-2 was cultured from the respiratory specimens (Kujawski et al., 2020). This report found some insightful data to back the origin SARS-CoV-2 and its natural history. Although infectivity is uncertain, highest levels of viral RNA were acknowledged during the early weeks of illness. These recent reports on CT value have an important suggestions to the clinicians, it should be anticipated that some of the infected patients may show aggravated symptoms during the second week of the illness onset.

3.2. Nucleic-acid testing workflow for SARS-CoV-2

Though, the RT-PCR method is being largely used for identifying SARS-CoV-2 infections by collecting respiratory swab samples in accordance with the recommendations designed by the World Health Organization (WHO). Samples from the upper respiratory tract can be collected through an oropharyngeal swab, nasopharyngeal swab, nasopharyngeal wash, and nasal aspirates (Pondaven-Letourmy et al., 2020). The swab samples from the upper respiratory tract are predominantly advised but samples from the lower respiratory tract are also recommended by the Center for Disease Control and Prevention (CDC), a national public health institute in the United States, particularly for patients suffering from coughing symptoms. Samples from the lower respiratory tract are not compulsory to be collect either from BAL fluid, aspirates from the tracheal region, or sputum (Thwe and Ren 2020). Sample collection from both of the BAL fluid and aspirates from the tracheal region have a greater risk of aerosol droplet generation and rapid spread to the sample collection environment (Sanyaolu et al., 2020). On the other hand, the possible viral-load is dependent on the number of days have passed after the onset of infection and the vulnerability of patient health (Zhou et al., 2020a). Thus, SARS-CoV-2 virus infections has to be consistently identified using sputum samples in addition to nasal swabs after 14 days of infection onset, whereas throat swabs are unreliable after 8 days of symptom onset (Pan et al., 2020c). A negative test result from the respiratory tract samples some time cannot be reliable indication of the infection due to viral load variability. Negative test results can be expected due to incorrect sampling and also in case of the low viral load, false negative test results may

be also anticipated in the latter stages of the pandemic owing to the mutations in the some of the viral gene sequences (Ai et al.;2020; Winichakoon et al., 2020). Winichakoon et al. has also recommended that the investigation using multiple test types is mandatory for the suspected patients, particularly for patients those shown some of the epidemiological evidence although well-adopted test results appears to be negative for samples collected from the upper and lower respiratory tract.

There is a need to integrate the nucleic acid detection workflow with other epidemiological evidence to improve clinical accuracy and throughput. Central workflows have to be proposed and implemented for RT-PCR test results in the established diagnostic settings. A three-step workflow was recently proposed by Corman et al. to advance the diagnosis of SARS-CoV-2 infections (Corman et al., 2020); the authors defined this three-step protocol for rapid screening, confirmatory test results, and discriminatory sample testing. To scale-up the diagnosis of a large number of suspected individuals, the first step screening has to be performed to trace all viruses related to SARS by targeting the E gene and different other regions in the gene. In the preliminary efforts, if the screening test is positive, targeted diagnosis moves for the RdRP gene as recommended to perform at least using two variants of probes and two variants of primers. If all test results appear positive or if any of the results are mixed, a final discriminatory test has to be performed using one of the designated probe for precisely diagnosing SARS-CoV-2.

Another unique workflow was proposed by Chu et al. for the screening and confirmation of SARS-CoV-2 test results (Chu et al., 2020). The authors suggested that the screening of test samples using primers for both an open-reading frame 1b (ORFlb) gene and the N-gene have to be used to confirm unclear test results. That report further clarified that a positive test result for the N gene primer with a negative test result for the ORFlb-gene would-be quit indecisive in arranged clinical settings. In such ambiguous situation, a protein tests based on antibody diagnosis or newly emerged protocols would-be further required for sequencing and confirmation of the test results. The important and recent report described an alternative workflow model having features of simplicity and rapidity for molecular detection of SARS-CoV-2 RNA with a higher sensitivity about 97.4%, wherein test samples were basically processed with heat for 5 min at a temperature about 98 °C prior to the mostly-used RT-qPCR technique (Fomsgaard and Rosenstierne 2020).

Throughout the pandemic SARS-CoV-2, economic aspects associated with diagnostics are multifactorial and complex to understand. Influence by administrative control, cost-effective concerns of workflow in clinical laboratory settings, technological advances, the needed investments to arrange laboratory devices, levels of R and D funding, in addition to this there is need for considerable high-end tests and enhanced database management (Jin et al., 2020), and the arrangement of IT solution, altogether has an important economic significance (Vandenberg et al., 2020). The two important but opposite significance for laboratory diagnostic activities have to consider prudently, the first one, how to manage huge rush over microbiological departments in addition to their diagnostic activities considering a significant increase in the number of suspected patients with SARS-CoV-2 (Dhama et al., 2020). The second one is the clinical laboratory activities those are not concerned directly to SARS-CoV-2 drops significantly, comprising, e.g., genetic sequence testing, when it adapts to the different or countryside facility model (Pereira et al., 2020). Moreover, when the lockdown was enforced, portability efficiency clogged (Rajan and Joshi 2020), delivery of surgical services impacted (Søreide et al., 2020), thus caused an immediate influence on the economy of the workflow managers and ultimately on the health-care providers. International Gemological Institute testing consortium has developed a dynamic training program for the SARS-CoV-2 testing team, proficiency assessment for those members involves in biosafety and assay workflow team responsible in the entire test workflow, whereas other members are likely to get trained specifically for the specific tasks they execute (Amen (2020)). In view of, there is mounting demand for molecular reagents and RT-qPCR diagnostic kits involve in the extraction and detection of SARS-CoV-2 RNA, at the same time there is the possibility of the global risk of scarcities (Feng et al., 2020).

Some challenges with the RT-PCR method has to be reported, including the limited availability of reagent kits owing to their large demand, lockdown, lack of facilities in rural cities, shortage of the infrastructure essential for RT-PCR, and some other limitations over accommodating high testing throughput. Another challenge with RT-PCR is that the test results rely on the existence of a measurable amount of SARS-CoV-2 test sample. RT-PCR method being not applicable for those patients who have been completely recovered after asymptomatic infection of SARS-CoV-2. Thus, SARS-CoV-2 infection is currently diagnosed predominantly using RT-PCR kits and patients can be also screened using chest CT and some other serological tests; however, each of these techniques has its own limitations.

3.3. *Computed tomographic scans for SARS-CoV-2*

On the onset of emergence, there was nonexistence or shortage in supply of RT-PCR kits, mere facilities for providing testing services, and the possibility of resulting false-negative results, thus, clinical personnels were suggested to use chest CT scans for clinical examination of SARS-CoV-2 infections severity (Yang and Yan 2020). Chest CT test can provide a detailed report on pathophysiology that could be useful to reveal the severity of disease evolution and detection (Yu et al., 2020b). A normal chest CT scan is a non-invasive technique that allows examinations of the infected patient's chest by performing with many different angles of X-rays collecting cross-sections in the form photographic images (Lee et al., 2020; Whiting et al., 2015). Such images need to be examined by expert radiologists to identify abnormal features that in the chest tissues, that helps with other diagnosis. The imaging features of SARS-CoV-2 patient's chest are not specific but vary depending on the viral load and severity of infection after the onset of severe symptoms. For instance, Bernheim and group revealed that CT findings frequently appears to be normal (55%) during the initial stages of the SARS-CoV-2 infection (0–2 days) (Bernheim et al., 2020); however, the diverse range of lung tissue distortions was observed at about 10 days of symptom onset (Bernheim et al., 2020). However, most predominant symptoms with the highest peak can be observed for SARS-CoV-2 patients including peripheral and bilateral grounds with glass-like opacity (chest regions with foggy opacity), such observations were most noticeable on and around 4 days after the onset of symptoms (Kobayashi and Mitsudomi 2013).

Likewise, evident observations like compacted lung tissues with consolidation of different features in addition to the solid, fluid, and some physical matter reported (Bernheim et al., 2020; Pan et al., 2020b). When SARS-CoV-2 infection severity progresses, some other types of observations like grounded glass-like opacities, bizarre paved-like patterns (paved stone with irregular shapes) appears (Pan et al., 2020b), in addition to swelling of lung tissues. (Bernheim et al., 2020; Pan et al., 2020b). Based on chest CT scan imaging observations, numerous retrospective studies have shown the technique has good sensitivity (85–95%) but a higher possibility for false-negative results as compared to the well-established RT-PCR testing kits (Fang et al., 2020; Guan et al., 2020). CT and RT-PCR mostly come in concordant, CT scans are applicable to detect early SARS-CoV-2 infection in those patients were negative by RT-PCR test (Xie et al., 2020), in those patients were without any symptoms, or earlier to the development of symptoms or even after symptoms got resolved (Ai et al., 2020; Inui et al., 2020).

Moreover, chest CT scan machines are expensive and require technical experts and radiologists. A recent consensus report made by international radiologist experts supporting the use of chest CT scan for diagnosis of SARS-CoV-2 patients particularly those are with poorest breathing status or in a resource-limited settings at clinical triage equipped for patients suffering from moderate to severe symptoms in

addition to those have high pretest possibility of SARS-CoV-2 (Rubin et al., 2020). The technique also cannot diagnose SARS-CoV-2 with high specificity, therefore, some of the recently developed technologies need to be adapted to simplify the confirmatory tests for SARS-CoV-2. Consequently, some guidelines provided by experts were against using chest CT particularly in screening or confirmatory diagnostic settings partly owing to similar radiographic images with other disease associated with influenza. Besides this, the potential limitation of using chest CT scan imaging has low specificity of about 25% to diagnose SARS-CoV-2 and another reason is that imaging features may be similar to those for other viral-caused pneumonia-like infections (Zheng et al., 2020b). Emerging artificial intelligence techniques useful for distinguishing between these observations may reinforce support in the direction of practice of CT scan in the diagnostic settings (Mei et al., 2020). A recent report developed AI algorithm and evaluated for the diagnosis of SARS-CoV-2 using chest CT scans applicable for data from a worldwide and from all sort of institution datasets (Harmon et al., 2020). This study showed the possibility of preparing robust models that can achieve accuracy up to 90% in diverse test populations, with retaining high specificity in other related pneumonia infections, and thus validated sufficient predictability to unnoticed patients.

4. Emerging diagnostics for SARS-CoV-2

At present, RNA detection of SARS-CoV-2 by BAL and nasopharyngeal swab samples, together with some blood testing parameters and chest CT scan, are the predominant tests for confirmation of suspected patients (Nuccetelli et al., 2020). Detection of any reported viral nucleic acids can be performed by real-time quantitative PCR (RT-qPCR) method in the approved test centers, under bio-hazard safety class II. However, RT-qPCR test kits are encountered to give some false-positive or -negative results, are thus, subject to swab sampling route and RNA extraction protocol, and there is the possibility with the virus, even though present in the patient, is not detectable within the mucous membrane of the nose–pharynx (Nuccetelli et al., 2020). In recent times, some claims are about a discrepancy between diagnostic efficiency of CT and RT-qPCR, the latter been identified as more sensitive. There was incidences of appearing false negative reports with molecular tests, sometimes there were forceful attempts of repetition of the same tests, frequent tests in clinically suspected patients and/or with CT scan pattern (Nuccetelli et al., 2020). A novel, molecular user-friendly tests are on the prospect for point-of–care (POC)/out of lab screening tests for SARS-CoV-2 RNA, using test samples without heating and extraction obliterate nuclease and cards that runs on clusters of regularly interspaced short palindromic repeats (CRISPRs), as a method DNA endonuclease targeted trans reporter with CRISPRs (Nuccetelli et al., 2020). On this context, constant debate is broke on discussing the significance of rapid serological assays those are based on detection of IgA, IgG, or IgM in plasma serum samples as anti-SARS-CoV-2 (Liu et al., 2020e; Okba et al., 2020), or in capillary blood, to make various decisions and clarify picture magnitude of the existing outbreak in each state or country, to support the final decision made by physicians and also to assess the need of scale of immunization (Nuccetelli et al., 2020).

Some of the POC tests, serological assays have been recently developed after the emergence SARS-CoV-2 pandemic, including rapid antibody immuno-chromatographic tests (RAICT), enzyme-linked immunosorbent assays (ELISA), POC-fluorescence assays, and chemiluminescence immunoassays (CLIAs) (Nuccetelli et al., 2020; Okba et al., 2020). Hundreds of different brands have been recently proposed to provide diagnostic kits and ready to approval and ship to the market, many of them are approved from European countries and few are being approved by Food and Drug Administration (FDA) (Abbasi 2020; Okba et al., 2020). Serological diagnostic tests are applicable to detect anti-SARS-CoV-2 antibodies against generates as responses to the viral antigens are yet to be widely used during the existing pandemic, however somewhat in a “POC” manner in private and public facilities (Okba et al., 2020); however, such diagnostics would be also as worthwhile as they were throughout the SARS epidemic of 2002 and recent studies have demonstrated that the IgG/IgM occurrence as anti-SARS-CoV-2 in diagnostically confirmed patients with negative results from RT-qPCR tests (Liu et al., 2020e). Serological diagnostic tests are more cost-effective than those of molecular tests based on nucleic acid detection (Dowdy et al. (2020a); Kubina and Dziedzic 2020), also require shorter testing time, thus screening at the large scale populations can be achieved than those of molecular tests, in case of medium to large size hospitals with automated instrumentation facilities employing ELISA and CLIA (Montesinos et al., 2020).

Though the RT-PCR method is a gold standard considered for the molecular identification of SARS-CoV-2 (Corman et al., 2020; Zhu et al., 2020a), its necessity of thermal cycler makes non-ideal for POC applications. Alternative emerging techniques based on exponential amplification, e.g. recombinase polymerase amplification (RPA), loop-mediated isothermal amplification (LAMP), exponential amplification reaction (EXPAR), rolling circle amplification (RCA), and exponential strand displacement amplification (E-SDA), each of them can be performed at particular temperature setting. These potential techniques can be used without thermal cycling and also have the potential for POC applications. High sensitivity provided by RPA and LAMP technique is achievable using PCR detecting nucleic acids with low copy number, while some of these techniques are incompetent to accomplish the higher level of sensitivity (Ganguli et al., 2020; Zhao et al., 2015).

4.1. Reverse-transcription loop-mediated isothermal-amplification (RT-LAMP)

Nucleic acid-based testing with isothermal-amplification is presently under readiness with their good accuracy and ease of diagnosis for SARS-CoV-2 infection. This method is named so because the technique gets conducted at a selected temperature condition and it does not require expensive or special laboratory settings to provide an excellent analytical sensitivity like RT-PCR (Craw and Balachandran 2012). The technique involves either helicase-dependent amplification or RPA, named as a loop-mediated isothermal-amplification (LAMP). Numerous institutions are now successfully investigating and developing LAMP technology combined with reverse-transcription (RT-LAMP) and it has also being evaluated clinically for their accuracy, rapidity and POC diagnosis service for SARS-CoV-2 suspected patients (Yang et al., 2020c; Yu et al., 2020a; Zhang et al., 2020f). The RT-LAMP technology involves DNA polymerase with different target primers those has to be identified while interacting with distinct target regions of the viral genome sequence. Herein, this process may also employ two different inner-primers also called as forward primers and reverse primers, and two different outer-primers in the four-primer setting. RT-LAMP technology can be highly sensitive and specific because it is applicable to many distinct primers (Notomi et al., 2000). In RT-LAMP diagnostic examinations, the test samples in the tube use to amplify the target DNA strands that would be detected with different strategies such as a colorimetric reactions, pH-sensitive dyes, or fluorescence assays using a fluorescent dye those specifically binds to the c-DNA strands (Mori et al., 2001). The RT-LAMP assay can be completed within 1 h at 60–65 °C and its analytically sensitivity is within a limit of detection of ~70 to 75 copies/μL, present in the test samples. RT-LAMP reactions are easy to visualize and simple to perform, with low background noise signals. The detection SARS-CoV-2 infections can be also performed at POC service centers located remotely without need of thermocycler (Notomi et al., 2000; Zhang et al., 2020f). The accuracy and versatility of the newly developed RT-LAMP method conveys that such developments are suitably applicable to curb the existing threat of SARS-CoV-2 spread, even without the sophisticated infrastructure and availability of advanced molecular biology machinery. However, limitations of the RT-LAMP method has to considered by investigators, finding the better ways to improve primers selection and optimization of assay conditions,

hopefully, these developing technologies would assist in tracing and detection of SARS-CoV-2 infections (Mori et al., 2001).

4.2. Isothermal amplification combined with CRISPR technology

Clustered regularly interspaced short palindromic repeats (CRISPR) based diagnostics tools emerged from the findings of microbial CRISPR and (Cas) proteins associated with CRISPR. Cas12 and Cas13 are two types of Cas proteins (Abudayyeh et al., 2017; Chen et al., 2018), which are particularly reported as useful for developing analytical platforms. Both Cas12 and Cas13 proteins can be guided by CRISPR RNA (crRNA) technology specifically targeted nucleic acid sequences, though the single-stranded regions of crRNA are complementary to the target (Chen et al., 2018). The mechanism of Cas12 and Cas13 bindings to the nucleic materials are different, Cas12 typically targets single-stranded DNA (Chen et al., 2018), whereas Cas13 targets specifically single stranded RNA (Abudayyeh et al., 2017). The targeting sequences for Cas12 are typically chosen immediately to be next to the protospacer adjacent motifs (PAM). Thus, PAM can facilitate the unwinding of the chosen dsDNA target and binding between DNA targets and crRNA. Such binding then induces conformational changes within Cas12 proteins and finally activates its collateral cleavage activity to make cleave at any ssDNA arbitrarily. However, other Cas13 proteins target specifically the ssRNA (Chen et al., 2018), and its process does not need any requirement of PAM-like sequence.

Feng et al. demonstrated the possibility of combining isothermal amplification with the CRISPR technology for the improving detection of SARS-CoV-2 (Feng et al., 2020). RNA of the SARS-CoV-2 is first to get extracted from suspected patient samples, then purified RNA samples need the reverse transcription to form cDNA copies and thus, gets amplified by isothermal techniques either RT-LAMP and RT-RPA. The cDNA amplicon generated are either mixed directly to the Cas12-CRISPR system or transcribed to ssRNA copies first and then mixed to the Cas13-CRISPR system (Broughton et al., 2020). That report further describes the process for Cas12 gets activated by dsDNA within CRISPR-specific targeted sequence (in red) to the cleaved reporters of ssDNA. Similarly, Cas13 proteins recognizes the RNA within CRISPR targeted sequences and thus ably cleaves its available reporters of RNA. The cleavage of the provided reporters generates fluorescence response within a fluorescence assay and in lateral flow assay, cleavage of reporter moves to the appearance of test line with clear signal.

Both Cas12 and Cas13 proteins can be successfully employed using the CRISPR technique combined with an isothermal amplification approach for the improved detection of SARS-CoV-2. The targeting sequence of CRISPR must be carefully considered and examined while designing specific primers applicable to the RNA amplification of SARS-CoV-2. The targeting sequence of CRISPR should be in between primers and within the amplicon, this way Cas protein-crRNA complex thus ably scans the amplicons and then specifically binds with the available complementary sequence targets. After the precise targeting, Cas proteins get activated and lead to complete their own collateral cleavage activity, thus non-specific cleavage of those ssDNA substrates by Cas12 and multiple ssRNA sequences by Cas13 proteins (Broughton et al., 2020).

A short single-stranded nucleic acid signaling reporter's ably labeled using fluorophore and their corresponding quencher can be also employed as substrates (Broughton et al., 2020; Gootenberg et al., 2017). The signaling reporter cleavage ably separates quencher from the fluorophore and establishes fluorescence signals that can be monitored in real-time or visually under the LED light source. Otherwise, such substrates can be timely labeled using biotin probe and fluorescein amidite (FAM) probe can be thus also used in the detection on SARS-CoV-2 test strips (Broughton et al., 2020). The lateral flow test strips coated with gold nanoparticles (AuNPs) and with anti-FAM antibodies are also effective to capture FAM fluorescein. Gootenberg et al. suggested that in the absence of the target sequence, the reporter remains unreacted and gets involved with streptavidin present on the control line, maintain AuNPs in a dispersion state which upholds red band, representing negative test report. However, in the presence of the RNA target of SARS-CoV-2, the amplicons activate Cas proteins and cleave the signal reporters thus facilitates the freed FAM and capped AuNPs to move to the test line, producing second red indicators, showing positive test report. Similar to any test strip reports, the signaling reporters used in excess helps to develop in a positive test report, owing to the excess intact reporters that are coated as well on the control line.

The important aspect of CRISPR-based detection technologies that should be highlighted is that the isothermal amplification sequences of products produces signals if the sequences exist correctly, which thus provides high analytical specificity than those of using pH indicators or fluorescent dyes owing to their non-specificity (Kellner et al., 2019). In contrast, assays using just Cas12 or Cas13 are not analytically sensitive enough in detecting viral RNA in samples (Kellner et al., 2019). In a short period of time, isothermal amplification accomplishes an exponential amplification, thus the combination of CRISPR technology helps to overcome limitations in terms of better analytical sensitivity. This method was ably detected 10 copies of RNA/microliter of RNA extract of SARS-CoV-2, both amplification and detection were achieved within 30 min of incubation, which was suggested to be useful for on-site analysis and POC testing.

RPA is also compatible to integrate with CRISPR technology for the reason that both requires identical optimal reaction temperature conditions. Patchsung et al. has previously reported a Cas13-RPA-mediated CRISPR approach, named "specific highly-sensitive enzymatic reporter unlocking system" (SHERLOCK) (Patchsung et al., 2020), and demonstrated its detailed protocol and application for the detection of SARS-CoV-2(Zhou et al., 2020d). The detection limit for SHERLOCK was reported about 10 copy/μL was demonstrated for the extracted RNA specimens and both amplification and detection protocols can be performed within about 1 h (Guo et al., 2020b).

Though compatibility of CRISPR and RPA technology has been already proven by using the DETECTR platform a single-step detection protocol for human papillomavirus (HPV), a similar single-tube Cas-RT-RPA assay protocol for SARS-CoV-2 detection is yet to be investigated and reported literature. A single-tube method has the potential to simplify the protocol, thus, leading this method as more suitable for on-site detection and there is also the possibility of reducing time and reduce the chances of amplicon contamination in the laboratory settings. However, all steps involved in such assays, including isothermal amplification, reverse transcription, and CRISPR-facilitated detection, need at least the involvement of the enzymatic system. The leading challenge of accomplishing all molecular reactions in a single tube system has to find cooperation in the reaction settings that allows used enzymes to work optimally.

Recent studies have revealed that the CRISPR-based technologies have great potential for developing POC test kits detecting SARS-CoV-2, although it has yet to be available for medical diagnosis. Few CRISPR-based technologies are ready to be applied to perform analysis of suspected patients. Broughton et al. reported aanalysis of 78 respiratory samples after extraction of RNA samples from 36 confirmed positives using DETECTR for SARS-CoV-2 with the achievement of 100% specificity and 95% sensitivity (Broughton et al., 2020). Still, more validation related studies are required before moving CRISPR-based techniques for on-site diagnosis and POC tests of SARS-CoV-2 if it can be performed with minimal instruments of sample treatment techniques those are user-friendly with the CRISPR diagnostic technologies.

Another prominent hybrid method based on DNA nano-scaffold is being recently developed, it also involves a chain reaction of extracted RNA from serum and saliva samples, it is now validated for rapid diagnosis of SARS-CoV-2, therefore, this method has the possibility to emerge as a good alternative to the existing RT-PCR method (Jiao et al., 2020). The DNA nano-scaffolds method involves the first construction of

self-assembly of self-satiating probes and nucleic acid strands. Subsequently, the SARS-CoV-2 RNA initiates hybridization of DNA probes like H1 and free H2 along with the nano-scaffold, thus, illuminated DNA nanostrings can be rapidly reached (Chauhan et al., 2020). This method is also advantages because the possibility of designing their probes locally and also temperature tolerance with the isothermal amplification, thus, can be projected to diagnose specific targets within short reaction time about 5–10 min and at mild temperature condition 15–35 °C (Bui et al., 2019). This report further describes several opportunities to meet together integrating a diverse range of optically active agents including fluorescent proteins, light-forming enzymes, nanoclusters, and metal chelates, and composite nanomaterials with a novel structural composite on designed DNA scaffolds are emerging with rapid pace (Yuan et al., 2014).

5. Emerging serological tests for SARS-CoV-2

Serological tests detecting SARS-CoV-2 specific antibodies in patient's blood are important at this time to: (a) trace suspected contacts; (b) stimulate serological surveillance at the national, regional, and local level; and (c) categorize those who might already have passed infection asymptomatically (Abbasi 2020). Let's assume there is defensive immunity, serological data can be utilized to choose who can be allocated at the workplace of infected patients, especially health professionals who work in the environments where can possibly be exposed to SARS-CoV-2. Mostly, *in vitro* diagnostic corporations avoid to report the details of the antigens involved in the assay, and thus it is challenging to recognize whether antibodies detected with such kits and procedures have a neutralizing effect or not on the infection, probably through the binding to viral S proteins of a receptor-binding domain (RBD) (Chen et al., 2020a; Tai et al., 2020). Tai et al. further reveal that the S protein of SARS-CoV-2 plays a significant role in viral and host cell interaction, fusion and intracellular entry, and thus, serves as an important target for developing antibodies, cellular entry inhibitors, and also development vaccines. Moreover, various serological tests can be developed and used retrospectively for autopsy diagnosis purposes and, also, such methods can be finally used collectively with recently established molecular tests successful owing to their higher diagnostic accuracy. In addition, on the horizon, serological diagnostic tests could play a significant role in improving the efficacy of the evaluation of any approved vaccines (Madore et al., 2010).

Though IgM antibodies formation may initiate as the viral particles reache in the respiratory tract, usually the timing of immunoglobulin generation from the 4 days on the onset of symptom from one to two weeks, thus limits its suitability in the acute phase proteins diagnosis (Padoan et al., 2020; Xiang et al., 2020a). However, it is also important to be pointed out that most of the molecular diagnostic tests signify an "instantaneous" scenario of probable virus infection, while serological tests show virus infection all through a wider phase of the transmissible process, whether or not it appears to a clinical significance. In case of serious symptomatic, the practical use of various serological tests in improving patients are recommended, wherein sampling of hyper-immune plasma should not be left off since antibodies responses may reach the competence to achieve deactivation of the virus population and avoid possible side effects of the prescribed treatment (Bloch, 2020). About this, FDA is directing international efforts toward developing blood sampling-based antibody-rich SARS-CoV-2 therapeutics. The investigations of clinical observations and humoral response profile using serological tests may contribute to account IgG serum concentration those are suitable for reveal details related to immune response, and also persistence time involved in any of the immunization trials.

Thus, diagnosis of viral proteins along with the detection of viral nucleic acids of viral particles provides complimentary evidence useful to confirm viral infection, also epidemiological and immunological status. Unlike nucleic acid methods based on exponential amplification that may result in false-positive owing to accidental amplification of contaminants nucleic acids, protein samples cannot be amplified directly or indirectly, thus reduces the possibility of false-positive results. Alternatively, if proteins cannot be amplified, detection of minute protein content is a challenging task of analytical chemistry, thus, there is the demand for ultrasensitive detection methods.

Several proteins of the virus SARS-CoV-2 are sequenced and identified, includes four structural key proteins named E, S, N, and M, and other 25 proteins (Feng et al., 2020). High-abundance structural proteins are known as the key targets in the diagnosis of SARS-CoV-2. There are two potential viral target proteins called S and N proteins that can be used in the diagnosis of SARS-CoV-2. N protein identified from SARS-CoV, which has now revealed for their high degree of sequence homology with the recently emerged SARS-CoV-2 (Grifoni et al., 2020a), this protein also identified to be produced in high concentration in both the SARS-CoV and the SARS-CoV-2 infected individuals (Kammila et al., 2008; Yarmarkovich et al., 2020). Although the N is a structural internal protein of virus particle, it was also found in the blood serum samples of infected patients in response to SARS-CoV (Li et al., 2005c). There are no reports suggesting the presence of N protein in human serum in response to SARS-CoV-2 even though the relationship of N protein of SARS-CoV and SARS-CoV-2 viral infections (Tilocca et al., 2020). Detailed examination of the SARS-CoV-2 transcriptome demonstrated that the transcript of N protein covers a great section and might indicates N protein to be with high abundance after SARS-CoV-2 infection. On the other hand, a quantitative analysis of the SARS-CoV-2 proteome has to be established. Alternatively, S protein can be well-preferred target for the diagnosis of SARS-CoV-2 since it is involved in the entry of the virus into the host cells and thus directly involved in contributing SARS-CoV-2 virulence (Ou et al., 2020). Though the presence of such proteins in serum samples does not certainly signpost active infection, Li et al. reported the possibility of detecting SARS-CoV N antigen for a long period of time about 25 days at the beginning of symptoms (Li et al., 2005c).

Presently certain affinity ligands existing headed for both the N and S proteins were revealed by independent studies. Monoclonal antibodies produce in the response to the S1 subunit of the S proteins (Goo et al., 2020) and also in response of the SARS-CoV-2 N protein have been well established (Che et al., 2004). Enzyme-linked immunosorbent assay (ELISA) kits are accessible worldwide allowing immediate diagnosis of antigen samples resulted in response to the SARS-CoV-2 (Carter et al., 2020). The ELISA kit was testified for recombinant produced N protein detecting successfully after addition to the human serum samples with attractive LOD values about 1 ng/mL of sample (Feng et al., 2020). The previous report suggest that the ELISA kit developed for the detection of SARS-CoVs N protein and successfully achieved concentration around 3 ng/mL in human serum (Che et al., 2004).

The key challenge that emerged while developing diagnostic tools that can successfully detect trace levels of viral proteins is that of the lack of availability of specific antibodies against each and every protein of the SARS-CoV-2 (Jiang et al., 2020). The alternative measure is to discover various affinity ligands other than those of the antibodies (Orooji et al., 2020). Song et al. described how to select aptamers that could target RBD of the SARS-CoV-2 of the S glycoprotein (Song et al., 2020). That report further revealed that two different DNA aptamers of having 67 nt (K_d = 19.9 nM) and 51 nt (K_d = 5.8 nM) in length. In another report by Zhang et al. also established peptide blocker agents that could interact efficiently with the RBD of the S glycoprotein (K_d = 47 nM) (Zhang et al., 2020b). These reports further suggested that both the peptide and the aptamers can be used in biotinylation, which can be also favorable to develop their possible applications as affinity ligand agents while establishing novel diagnostic assays. The earlier invention of synthetic peptides or aptamers proficient in the production of antibodies can be responsible for the development of improved user-friendly technologies. Aptamers were used in targeting the SARS-CoVs N protein, those have been proposed recently to be modified to diagnose SARS-CoV-2s N protein (Chen et al., 2020d). Although N protein found

identical about 90% for both SARS-CoV and SARS-CoV-2 (Feng et al., 2020), thus cross-reactive possibilities cannot be denied with SARS-CoV, however, is not a major issue as currently lack of SARS-CoV spread. SARS-CoV-2 proteins N, E, S, and M share 70–95% sequence similarity with proteins of SARS-CoV and at least about 30–50% with the MERS-CoV (Chen et al., 2020d). Thus, epitope relevant to each of the viruses can be carefully preferred to help to identify possible vaccine targets and reduce chances of cross-reactivity from the specifically selected proteins during the assay procedures (Ahmed et al., 2020).

Viral protein-based diagnosis involves both viral antigens and antibodies that generates from the immune response to viral infections, thus can also be castoff for diagnosing SARS-CoV-2 infections. However, changes in viral-load during the infection time-course for SARS-CoV-2 could make the diagnosis challenging task for these methods owing to variations in concentrations of viral such proteins. In particular, high viral-load observes in salivary samples during the first week of infection and onset of severe symptoms, however, such viral load scenario can gradually decline over time course of infection (To et al., 2020). Antibody-based protein tests are known to be advantageous for post-infection surveillance measures implemented to curb SARS-CoV-2 crisis. On the other hand, antibodies forms in response to the SARS-CoV-2 infections, thus offers longer window of opportunity for post-infection detection and accurate data collection. Potential challenges for the developing such diagnosis for high throughput screening and accurate serological diagnosis is the likely experiencing cross-reactivity among those antibodies produced in response to the SARS-CoV-2 infections and also to those formed in response to other types of coronaviruses. Plasma samples obtained from completely recovered SARS-CoV-2 patients are reported for the S protein that to exhibit a high incidence of cross-reactivity reactions among SARS-CoV and SARS-CoV-2 as well as with some of the other coronaviruses (Lv et al., 2020).

At present, some of the serological tests are in use for diagnosis of SARS-CoV-2-specific antibodies after series of trials, validation, and approval from the administrative control (Zhang et al., 2020e). Zhang et al. reported that enzyme-linked immunosorbent assay (ELISA) ably detects immunoglobulin M and G (IgM and IgG) produced in serum samples of SARS-CoV-2 infected patients. This approach also uses the Rp3 nucleocapsid protein specific marker for SARS-CoV-2 that has amino acid sequence homology of about 90% with some of the other SARS-family coronaviruses. In this method, recombinant viral proteins allows first to adsorb onto the 96-well Petri plates and the excess proteins get washed out followed by dilution of human serum added to the plates needs to be washed again. Functionalized anti-human IgG in addition to the horseradish peroxidase enzyme allowed to perform reaction and bind to the target serological marker, finally, those plates used in this test have to be washed again and treated with substrate 3,3′, 5,5′-tetramethylbenzidine (Harpaz et al., 2020). Enzyme horseradish peroxidase reaction with substrate provided can be used for developing colorimetric sensors that are easy to monitor with a naked eye and microplate readers (Zhao et al., 2020). The extracted IgG anti-SARS-CoV-2 samples present in the test tube reacts with a horseradish peroxidase enzyme and forms sandwich between the antihuman IgG probe and the adsorbed nucleoprotein, thus produces accurate result. A detailed protocols are still under development for IgM testing, which is similar however when (anti-human, IgM) get fixed onto microtiter plate surface, anti-Rp3 nucleocapsids perform as a target probe (Udugama et al., 2020). Serological markers from the SARS-CoV-2-confirmed patients are being tested and also can be confirmed using the RT-PCR method, it was revealed that the levels of such antibodies increases during the initial 4–5 days of onset of symptoms (Zhang et al., 2020e).

Furthermore, 80% and 50% of SARS-CoV-2-positive patients produces IgG and IgM in the first few days after infection, respectively, but these values may increases up to 100% and 81% on after day 5 of onset of symptoms (Huang et al., 2020a). Antibodies were also being detected in different samples from the infected patients, including blood, respiratory tract, and fecal samples (Wölfel et al., 2020). The sensitivity for ELISA IgG, ELISA IgM, and colloidal gold-immunochromatographic assay (GICA-IgM, and GICA-IgG) diagnosis was found to be above 80% (Xiang et al., 2020b). There were no significant differences in sensitivity between GICA and ELISA (IgM, IgG) tests and these techniques shows negative signals for healthy controls, thus, specificity is almost 99% (Xiang et al., 2020b). The GICA assay used for the diagnosis of IgM and IgG antibody proteins specific to the SARS-Cov-2 has litter lower sensitivity but higher specificity, showing great potential for rapid detection of SARS-Cov-2 (La Marca et al., 2020). However, further studies should be performed to assess this method in diverse populations and different clinical settings (Shen et al., 2020).

Besides the recent reports, there are many other options that are emerging based on protein or clinical markers that are currently in the phase of research and development (R and D) that would be applicable to perform diagnosis of SARS-CoV-2 suspected individuals. Guan et al. has recently reported that the SARS-CoV-2 patients show elevated levels of both the serological markers include C-reactive proteins and a fibrin degradation product (D-dimer) in addition to the decline in levels of white blood cells (lymphocytes, blood platelets, and leukocytes, etc.) (Guan et al., 2020). Furthermore, the major challenge with this approach lies in the use of low levels of biomarkers, which are similar to the levels observed in other illnesses (Morales-Narváez and Dincer 2020). In this scenario, a multiplex detection test based on both antibodies and molecular markers can not only improve the sensitivity but may also improve specificity to diagnoseSARS-CoV-2 virus-infected individuals. In future studies, there is a need to compare diagnostic platforms, such as ELISA and magnetic chemiluminescence enzyme immunoassay (MCLIA) (Long et al., 2020a), which can be widely used for diagnosis without involving test centers to fulfill different practical needs including personalized home testing, large population screening, and alternative POC solutions in diagnostics (GeurtsvanKessel et al., 2020). Long et al. succesfully compared three different platforms, which can be widely used in most of the diagnostic laboratories (some rapid tests, four ELISA tests, and high throughput CLIA assay, which are appropriate to address essential needs: for customized home testing, as a complement to molecular diagnostics and suspected population screening.

So far, emerging antibody detection assays are still away from satisfactory results. The lateral flow assay based on metal nanoparticle (paper test strips) are attractive options for routine POC diagnostics and rapid sensing of IgM/IgG antibodies. Though it can provide rapid results within 5–20 min, it gives simply binary information (yes/no) with very minimum sensitivities. Consequently, such methods are not appropriate to cast-off for the qualitative estimation of the recovering human serum samples (Tan et al., 2020b). Alternatively, conventional ELISA tests can results sensitive and accurate analysis, but it needs expensive and complicated instrumentation and also a longer assay time about ~3 h (Ju et al., 2020; Wang et al., 2020a). Manifold dilution factor is mandatory to perform the serological tests for S1-specific IgG of SARS-CoV-2 in a given dynamic narrow range about <2 orders of magnitude, these factors increases the cost and decreases the throughput of the assay. Moreover, conventional ELISA is not able to measure the real concentration of the flowing IgG of anti-S1, thus, quality control practices for convalescent serum make harder owing to lack of having an internal calibration standard (Ju et al., 2020).

Microfluidic in diagnostic technologies are developing extensively in addition to their establishment in other applied fields (Chin, 2011; Han et al., 2013; Ng et al., 2018). Microfluidics-based technologies are appropriate to integrate the preparation of samples, reaction conditions, and detection steps into the preparation of miniaturized chips. Microfluidic technologies offer several advantages: (1) high throughput, portability, multiplex, and automation (2); it enables rapid, sensitive detection, ease of quality control, (3) it also saves requirement of reagents and cuts the testing cost (Ye et al., 2019).

In recent work, Tan et al. presented technology of microfluidic chemiluminescent based on ELISA for a portable quantitative, rapid about 15 min, and highly sensitive diagnosis for IgG specific to the S1 of the SARS-CoV-2 (Tan et al., 2020c). That report first considered four different traits of chimeric humanized IgG monoclonal antibodies and also described an appropriate candidate (D006) having high specificity and binding affinity to SARS-CoV-2s1 protein, it subsequently served as the accurate calibration standard for IgG of S1anti-SARS-CoV-2 in given serological tests. Furthermore, this report also revealed the ability of the developed microfluidic portable technology to demonstrate ultrasensitive detection of N and S1 protein antigens for SARS-CoV-2 with a low LOD value about ~10 pg/mL within 40 min of reaction for spiked serum samples.

As a result, the monitoring of specific immunoglobulin G/M, IgG/M antibodies, and corresponding antigens is an easy procedure, rapid, reliable, and handy approach for diagnosing SARS-CoV-2 infections as well as appropriate for establishing large-scale and efficient screening of suspected population at POC centers (Seo et al., 2020). The diagnosis of IgG and IgM in serum samples or whole blood has been established as a reliable technique to diagnose SARS-CoV-2 with high specificity and sensitivity (Cai, 2020; Li et al., 2020b). Furthermore, detecting antigen proteins of SARS-CoV-2 from swab samples in nasopharyngeal area also exhibited exceptional advantages for performing clinical testing (Seo et al., 2020). To encounter the challenge of the worldwide epidemic, there is a need to develop POC microfluidic platforms compatible with fluorescence detection analyzer, multiplex immunoassays, and diagnostic microchips for detecting different biomarkers (IgM, IgG, and antigen).

Some other reliable diagnostic techniques for protein detection are needed. Matrix-assisted laser desorption/ionization (MALDI-MS) equipment exists in most of the clinical diagnostic laboratories, which are mostly being used for the identification of fungal and bacterial infections. Recent reports proposed to influence the POC use and versatility for large-scale detection of SARS-CoV-2 testing using MALDI-MS. Such MALDI-MS assay does depend on the reference spectra collected for the particular pathogen and their bioinformatics database analysis while performing identification of strain with high-specificity and high-sensitivity with the help of proteomic profiling and database analysis. Such approaches are well established in most of the countries for daily diagnostic practices for the detection of common infections caused by yeast, fungal, and bacterial. However, proteomic spectral libraries are yet to be publicly available for the identification of SARS-CoV-2 using MALDI-MS assays. A recent report has firstly assimilated mass spectra of MALDI for samples from nasal swabs was tested for SARS-CoV-2 with RT–PCR and also examined then using machine learning approach (Nachtigall et al., 2020) with the MALDI mass spectra method to systematize assay procedures for SARS-CoV-2. Those nasal swabs samples first acquired for RT–PCR was also found useful for getting MALDI mass spectra without any sample purification protocol. Thus, the sample tested positive for SARS-CoV-2 can be also suggested to use in the follow-up the confirmatory test after the gold standard RT–PCR. Matching MALDI/ML and RT–PCR results reported, it with good concordance, thus, was acceptable for a performing alternative diagnostic method (Nachtigall et al., 2020). That report also proposed a systematic diagnostic protocol that could be applied through a level system integrating most of the robust ML prototypes, which could be also rapidly authenticated in any new laboratories, while being accepted as a rapid screening assay diagnose SARS-CoV-2 infections.

Validation experiments against gargle/saliva spiked samples with the cultivated virus have to be completed since a recent study specified that sensitivity was near 99% for determining S1 peaks particularly as an indicator of SARS-CoV-2 virus infection (Iles et al., 2020a). Straightway comparison of the RT-PCR testing with MALDI-ToF MS diagnosis of SARS-CoV-2 in clinically approved samples are desired. However, the MALDI-ToF MS method is not recommended for stored samples particularly pharyngeal-nasal swabs as they have to be either been inactivated by heat (heating the viral proteins and envelope membrane) or deposited in either SDS or Triton storage media which helps to suppress ionization and correspondingly abolishes the viral envelope materials. If there is a lack of freezer facility for prior-collected gargle/saliva samples to perform RT-PCR examination, those are already known, and in that case, only prospective testing is possible. Several of the researchers are effortlessly working with numerous other researchers collecting gargle/saliva test samples and process as a demo where a PCR testing for selected swabs can be simultaneously processed (Iles et al., 2020b; Rocca et al., 2020). Such research groups have to analyze different samples also using their MALDI-ToF instrumentation with necessary reagents involved and by their all possible technical support. As comprehensive research efforts may assist to make an analysis of all possible other notable markers by this technique, thus it may provide further essential medical information including the possibility of other viral infections and the degree of immune responses both humoral and mucosal.

Another method called Terahertz (THz) is a plasmon-based procedures are great for use as highly sensitive field analysis and detection, but exhibits the challenges of frequency tenability limits designed for fixed structures (Deng et al., 2019). THz-based methods however are showed strong potential to use as a molecular tool for a variety of analytical applications owing to its unique nature, including medicine, security, quality control inspection, and many more (Cai et al., 2014; Clery 2002; Kulesa 2011; Nakajima et al., 2007). The major hindrance in the consideration of THz uses is that the THz waves to the longer wavelength is relative to the ultraviolet, visible, and infrared light. The THz wavelength scale about 300 μm at 1 THz deters thorough studies of minute samples since the power expected by those samples considerably loses as the sample size reduces below to the sub-wavelength scale. This problem is largely critical for probing micro-, nano-scale objects such as nano-materials, cells, as well as biomolecules molecules, which is thus hindering the competence and utility of THz based diagnostic measurements. This concern needs to be highlighted for their importance of developing highly effective THz wave coupling and concentration with such nanoscale samples.

The effect of geometrical parameters is suggested to optimize the sensitivity of the devices, including a refractive index of substrate and antenna width to improve sensitivity. For example, for a better sensitivity, it is important to reduce the operational refractive index located at the gap area, which can be established with substrates having a low refractive index and essentially free-standing films (Tenggara et al., 2017). Thus, it is well-known that the sensitivity of devices can be considerably improved in relation to decreasing at the gap width due to the improved field localization (Park et al., 2015; Seo et al., 2009). Recently, nanoscale-gaps were prepared using metamaterials with nanolithography techniques to achieve sensitive detection of viruses having more than 100 nm (Park et al., 2015). However, such an approach is time-consuming and expensive to construct devices with nano-scale structures by using traditional lithographic techniques; thus, advanced techniques prerequisite accomplishing localized fields precisely. Consequently, it would be desirable to integrate novel functional nanomaterials that having inherent nanoscale sizes for example nanowires having one-dimensional. Recently, network-like films made up of nano-scale materials for example reduced graphene oxide, single-walled nanotubes, and silver nanowires have appeared as a potential alternatives for developing THz optoelectronic devices (De et al., 2009; Kim et al., 2013). Furthermore, solution-based procedures involve to practice relatively low-cost spin-coating approaches, in particular, AgNWs exhibit also exhibits superior optical and electrical properties, which makes them interesting to develop applications in THz based optoelectronic devices (2017); however, biomedical sensing applications with improves sensitivity are yet to be addressed.

Considering epidemiological concern, it would be important to examine the host-specific virus relationship, together with a correlation of immune-protection, includes a set of viral pathogen-specific

neutralizing antibodies that limit the SARS-CoV-2 infection (Ju et al., 2020) and correlate as immune dysregulation, such as over-expression cytokine that may increase the susceptibility to symptoms (Huang et al., 2020b; Sepe et al., 1981). Immune response factors, those related can constitute a fundamental immune signature associated with viral disease and various other elements of which do exist in other pneumonia-like infections and/or immune-pathologies, together with however not limited to those of immune response factors causes by some other animal and human coronaviruses (Saif 2004).

However, the viewpoint on a consensus SARS-CoV-2 immune signature has been challenged our scientific aspects by the viral infected patients heterogeneity, age, span, ethnicity, gender, clinical performance, and core symptoms of infection illness (Pan et al., 2020a; Saif 2004). Therefore, the increase of specific antibodies and T cells to SARS-CoV-2 (Grifoni et al., 2020b; Ju et al., 2020; Sekine et al., 2020; Weiskopf et al., 2020) have been identified variably set against the portrayal of immune-deficiency and lymphopenia (Blanco-Melo et al., 2020; Kuri-Cervantes et al., 2020), affiliated with the transfusion of infected patients with neutralizing antibody-rich convalescing plasma (Duan et al., 2020a); and metaphors of cytokine storms and neutrophilia (Moore and June 2020) (Huang et al., 2020b), for which immune-suppressants those are been recommended.

Admitting huge complexity immunogenic factors, we however thought that there is the existence of a core immune signature, analogous to the measurements of extremely diverse biomolecules those make signature responses to the infection and vaccination (Sobolev et al., 2016)or either displays alike dysregulation of immunological factors in a severe symptomatic stage (Ghnewa et al., 2020).

Laing et al. determined thyroid peroxidase and thyroglobulin autoantibodies in the serum sample using the electro-chemiluminescence assay by analyzer cobas 6000, after receiving recommendations from the Roche diagnostic manufacturer (Laing et al., 2020). That study also performed longitudinal analyses for treated patients at hospital suffering from SARS-CoV-2, it was revealed a vigorous, prevailing immune response, and it clearly helps to segregate patients from normal, including recovering seropositive patients. The immune response includes numerous discrete adaptive and innate traits, comprising changes in dendritic cells and B cell together with severely altered phenotypes of T cell those may perhaps undermine immunity of immune-protective T cell (Grifoni et al., 2020b) (Grifoni et al., 2020b). Furthermore, remarkable correlations have been currently appeared for example that related to the elevated interferon-inducible protein 10 (IP-10) and basophil pool depletion.

Some of such traits occur in other settings too, concerning but not limited to the immune-protective non-lower respiratory tract SARS-CoV-2 infection, sepsis and vaccination (Ghnewa et al., 2020; Sobolev et al., 2016). Alternatively, those mentioned and other traits together have can be composed with a core SARS-CoV-2 immune signature, while clinical characteristics detectible in other circumstances pool to constitute core symptoms of SARS-CoV-2. There is a need for providing a database on core immune signatures in open dataset maintained by online websites or portals like www.immunophenotype.org, in that way easing autonomous validation at different settings. However, the immune signature's comprehensive applicability have been suggested in the various reports for numerous of those traits in other SARS-CoV-2 research groups (Guo et al., 2020a; Liu et al., 2020b; Vabret et al., 2020; Wilk et al., 2020; Zheng et al., 2020a), though some other traits e.g., eosinophil, neutrophilia depletions, and natural killer cells composition may also support to be more diverse (Kuri-Cervantes et al., 2020; Wilk et al., 2020).

The core immune signature does offer several potential opportunities in developing rapid and accurate molecular diagnostics also by identifying matches with new settings, traits consist of upregulation of interleukin-8, depletion of $CD5^+$ B cells, activation of plasmablast, reduction of human leukocyte antigen-DR isotype expression by monocytes and subset-discerning to the T cytopenia (Ghnewa et al., 2020) (Cunha et al., 2009; Dunning et al., 2018; Ferreira da Mota et al., 2018). Thus, such accounts may help in the interpretation of shared pathogenesis and also may offer models for specific infectious disease management, also as illustrated by the interleukin-8 capacity to increase T cell competency in conditions like sepsis and also in SARS-CoV-2 as clinical trial report described on website portal (https://clinicaltrials.gov/ct2/show/NCT04379076). Some alternative traits, including pre-dendritic cells, basophil and $V\delta2^+$ T cell depletions those are mostly-related, and not evidently shared with non-SARS-CoV-2 lower respiratory tract infection and/or any other infectious diseases, thus, may provide tool to better understand and to track unique markers of SARS-CoV-2 pathogenesis (Varadé et al., 2020).

A possibly practical opportunity can be offered by the persistent triad of IP-10, IL-6, and IL-10 and some newly explored traits if quantified in early stage may help to diagnose accurately but also anticipate the severity of infection progression and need for admitting in hospital (Laing et al., 2020b). Potentially these traits can be used in performing routine clinical diagnostic tests to aid initial risk-based delamination of confirmed patients, which would be of foremost advantage when the healthcare system is over-stressed. Moreover, precisely targeting storm or depletion of SARS-CoV-2 specific inflammatory mediators may aid to strain therapeutic benefit, more probably contributing to the execution of therapeutic treatments (2020a). Although involvingthe application of this triad, the core immune signature has to be explored for a broad-base of cytokine storm experienced in most of the prevailing SARS-CoV-2 infections.

The essentially consistent production of RBD-specific IgG after SARS-CoV-2 infection indicates that patients may have immune-competent possibility to limit viral infection. However, the supposedly high affinity of SARS-CoV-2 cellular entry may be partly immune-evasive (Shang et al., 2020a), growing the dependence on the mechanisms eradicating viral-infected cells. In this regard, the burden of viral infection correlates rather with natural killer cells and $V\delta1^+$ effector cells, which both are involved in immune responses to the antiviral treatments (Agrati et al., 2001; Biron et al., 1989; Lafarge et al., 2001). Furthermore, immune-protective possibly was proposed by activation of multi-faceted T cell, together with upregulation of human leukocyte antigen DR isotype and expression of CD38 that was described specific T cells for SARS-CoV-2 (Sekine et al., 2020). Current and future revelations would portray SARS-CoV-2 virus-specific T cell responses, but their reports were applied to establish a profound T cell phenotypes of SARS-CoV-2 that evident frequency on either depletion or cycling, was almost indeed exceeded occurrences of virus-specific immune cells (Grifoni et al., 2020b; Sekine et al., 2020). Certainly, extensive dysregulation of T cells may weaken immune-protection mediated by T cells mostly as it significantly affects memory T cells wherein adult individuals get perhaps more dependent than those of children's owing involution thymic system as reported in previous studies (Lyu et al., 2020; Salam et al., 2013). Such immune cells may take account of being possibly protective but the elderly population certainly suffers from metabolic diseases such as diabetes and hypertension, thus, commonly involving memory T cells apparently remained well-informed against common cold infections like pneumonia as well as coronaviruses (Grifoni et al., 2020b). Worth mentioning, seropositive persons those had newly recovered without any treatment, therapeutic interventions, or hospitalization confirms lingering traits with activation of T cells (Li et al., 2020a).

Postmortem reports of SARS-CoV-2 victims helped to reveal lung infiltration specifically myelomonocytic (Carsana et al., 2020) those may partially justify the loss of plasmacytoid dendritic cells and/or basophils from the blood, with that of latter develop implication while lung tissue gets repaired (Crivellato et al., 2010) and while coagulation regulation (Merle et al., 2015; Swystun and Liaw 2016) that may be useful to create recurrent thrombotic events also in SARS-CoV-2 infections. Similarly, T cytopenia may replicate trafficking, e.g., in the lung tissues, though merely diverse infiltrations of lymphocytic were shown after postmortem analysis (Carsana et al., 2020; Crivellato et al.,

2010), and T_{EM} residual cells does not clearly show any significant expression of chemokine receptors.

As an alternative, T cells may perhaps be in a hyper-activated form, develop susceptible to apoptosis (Chen and John Wherry 2020). Yet again, some other postmortem reports have shown white pulp of splenic, and pulmonary and necrosis of hilar lymph nodes with the depletion of T cells in a setting of activation of B cells (Prilutskiy et al., 2020). However, depletion of both $CD4^+$ T_N and $CD8^+$ T_N seems challenging to characterize wherein cell death or homing gets induced by activation, those may perhaps reveal an active differentiation of central memory T/effector memory T cells; $CD8^+$ T cells mediated fratricide event or hemo-phagocytosis (Yang et al., 2020b). It may also be expected that the diverse metabolic profiles from distinct T cell subgroups may possibly render them specifically vulnerable to dysoxia, hypoxia, or particular inflammatory molecular markers that are yet to be identified.

A noteworthy observation of upregulation of interferon gamma-induced protein 10 (IP-10) a near-universal (Coperchini et al., 2020), assumed to depletions of T helper cells 1, T helper cells 17.1, and plasmacytoid dendritic cells, and cells those might normally be causes of both type-I and type-II interferons those acts as the key inducers of IP10. Though IP-10 levels exhibited some relationships with interferon-gamma (Chen et al., 2020c), and even though some confirmed patients showed a high level in addition to the transient interferon-gamma levels, IP-10 may perhaps be also boosted by some other factors and virus-associated mechanisms. Certainly, an elevated levels of IP-10 can be specifically characterized as reported for SARS-CoV (Huang et al., 2005) and MERS-CoV (Shin et al., 2019), those enters to the T cells via CD26, an as reported ecto-peptidase do control the activity of IP-10 (Casrouge et al., 2012). Possibly, dysregulation of chemokine is known for a core component of coronavirus infections and pathogenesis, perhaps interfering with ordered chemotaxis of immunocyte and thus, contributes to a setting wherein coronaviruses reportedly thrives (Ye et al., 2020). Probably relevant to this concern, three kinds of receptor genes for chemokine lies within a region related with a severe respiratory SARS-CoV-2 disease susceptibility (2020c).

In conclusion, the multi-faceted immune signatures of SARS-CoV-2 may convey a solid basis for addressing many of the current research and clinical inquiries, together with whether or not individual immunological traits, can be considered discretely or collectively together, are institute or significances of progression of SARS-CoV-2 disease. Without a doubt, our current understandings on SARS-CoV-2 disease and its immune response are rapidly evolving, appearing with the rapid pace, intensity, and with the global range of R and D activities. The global scope would provide great relevance to the context for immediate and depth assessments on the SARS-CoV-2 specific immune signatures in various settings, improving the predictions of agreement considerate and collective activities to enhanced curb infectious diseases and thereby to reduce the life-threatening possibilities caused by SARS-CoV-2 infections.

6. Point-of-care services

The SARS-CoV-2 diagnostic research with the urgency mostly focused on nucleic acid molecular diagnostic kits, protein test kits, and testing at POC service centers are being establishes. The long-term priority also needs to be seriously considered to integrate different tests into multiplex panel systems (Zhu et al., 2020b). Serological tests using protein-based test kits are now being deployed in addition to nucleic acid-based test kits to improve surveillance efforts. Protein-based diagnostic tests can detect asymptomatic infections following complete recovery of patients unlike nucleic acid-based test kits, which have limitations in detecting those patients being already recovered patients. Protein-based test kits may, thus enable clinicians to screen and contact-trace both infected and recovered patients, which increases the possibility of estimating the entire SARS-CoV-2 infected population. POC service centers can make testing SARS-CoV-2 infections more cost-effectively and portable and enables the diagnosis of patients far from centralized facility centers in urban community health centers, thus reducing the burden on clinicians, as reported for POC in cancer detection (Mahmoudi et al., 2020).

POC test approach allows diagnosing suspected and infected individuals without transporting their test samples to the central facilities, thus allowing detection of SARS-CoV-2-infected patients at local community centers, without sophisticated laboratory facilities. A lateral flow SARS-CoV-2 antigen assay is under development for POC diagnosis of infected patients (Xiang et al., 2020b). Lateral flow commercial assays are based on the membrane or paper-like strips coated with two lines, with antibody-conjugated gold nanoparticles on a line and captured antibodies to the other line. The sample (either blood or urine) has to be exposed to the membrane where test proteins from the sample are strained across the test strip the mechanism is based on capillary action. After the test sample crosses the noticeable first-line wherein antigen proteins do bind with the target antibody conjugated onto the metal nanoparticle like gold (AuNPs) and the resulting complex travels together throughout the used membrane. Consequently, the test sample reaches onto the second line where the final complex interacts with the captured antibody proteins and becomes immobilized; thus either blue or red line appears visible to the naked eye. Monodispersed AuNPs exhibit a dark red color, while aggregated one in a solution exhibits a dark blue color owing to the surface plasmon resonance coupling band. The recently developed lateral flow antigen assay for SARS-CoV-2 was found to have an analytical sensitivity, accuracy, and specificity approximately about 60%, 69%, and 100%, for IgM antibodies, respectively, and for IgG antibodies for about 81%, 86%, and 100%, respectively (Udugama et al., 2020). Thus lateral flow assay can also be integrated with nucleic acid testing. A previous report on the RT-LAMP test results suggests that this approach could be also incorporated with a lateral flow assay system for the diagnosis of MERS-CoV (Huang et al., 2018). Thus, such assays are suitable for single-use tests and also have lower analytical sensitivity than RT-PCR. Therefore, researchers need to develop a variety of techniques that can be used in signal amplification including the assembly of small antibody functionalized AuNPs and thermal imaging systems that would improve the assay readout signal (Spengler et al., 2015).

Another important method that can be considered for a POC system is a microfluidic device-based method. These devices involve a small-sized chip imprinted with reaction cavities and micrometer-sized passages. The chips are getting prepared using common materials such as glass, poly-dimethyl sulfoxide, as well as functionalized paper to separate and mix some liquid samples using a mechanism of capillary, vacuum, or electro-kinetic forces. The main benefits of the microfluidic system include the need for small sample volume, rapid diagnosis, miniaturization, and good portability (Foudeh et al., 2012). Progress on techniques using microfluidic platforms is necessary to simplify the detection of biomarkers, amplification, sample preparation, and fluid handling to achieve POC diagnosis as well as high-throughput multiplexing. A microfluidic system and smartphone dongle-based attachment was suggested to detect different types of antibodies after the onset of infection by successively moving reagents on a sample holder (Laksanasopin et al., 2015). Laksanasopin et al. reported that this alternative platform has a clinical sensitivity of about 100% and a specificity of about 87% for the human immunodeficiency virus (HIV). The possibility of adapting technologies based on microfluidic devices to diagnose the target RNA or proteins of SARS-CoV-2 should be constantly investigated.

Herein, we summarized some other diagnostic tools that would be appropriate in terms of clinical practicability. Table 1 is prepared to presents a wide-ranging list for various emerging methods which would be appropriate after improvement for accurate and specific detection of SARS-CoV-2. Such alternative platforms' diagnostic applications would also be introduced by R and D teams and academic laboratories, including paper-based colorimetric systems, electrochemical devices, and surface-enhanced Raman scattering systems (SERS), MALDI, and

Table 1
Currently emerging wide-range of diagnostics tools applicable for SARS-CoV-2.

Platforms	Biomarker	Technology	Sample Source	Contagion	Sample Tested	POC (Y/N)	References
CRISPR	Nucleic acid	RT-RPA	Nasopharyngeal swabs	–	384	Y	Kellner et al. (2019)
CRISPR	Nucleic acid	RPA	swine serum	ASFV	110	Y	Wang et al. (2020b)
LAMP	Nucleic acid	LAMP	Throat swabs	H5N1 influenza	53	N	Imai et al. (2007)
LAMP	Nucleic acid	RT-LAMP	Nasopharyngeal aspirates	Respiratory syncytial virus	59	N	Shirato et al. (2007)
RPA	Nucleic acid	RT-RPA	Fecal and nasal swabs	Bovine coronavirus	30	N	Amer et al. (2013)
RCA	Nucleic acid	Rolling circle amplification	Serum	Hepatitis B virus	7	N	Martel et al. (2013)
NASBA	Nucleic acid	Real-time NASBA	Nasal swabs	Respiratory viruses	138	N	Wat et al. (2008)
Quantum dot	Nucleic acid	Barcode assay	Serum	Hepatitis B virus	72	Y	Kim et al. (2016)
Smartphone dongle	Protein	ELISA	Blood	Sexually transmitted diseases	96	Y	Laksanasopin et al. (2015)
Paramagnetic bead	Protein	Magnetic biosensor	Serum	Dengue virus	12	N	Aytur et al. (2006)
Magnetic bead	Nucleic acid	Magnetic	Stool	*Helicobacter pylori*	17	N	Nilsson et al. (1996)
Magnetic bead isolation	Whole bacteria	Magnetic separation	Synovia	*Staphylococcus aureus*	12	N	Bicart-See et al. (2016)
SIMOA	Protein	Digital ELISA	Serum	–	30	N	Rissin et al. (2010)
ELISA	Protein	ELISA	Serum	Avian influenza A	30	N	Rowe et al. (1999)
Rapid antigen test	Protein	Lateral flow	Serum	Zika virus	117	Y	(Bosch et al., 2017)
Biobarcode assay	Protein	DNA-assisted immunoassay	Serum	Prostate-specific antigen	18	N	Thaxton et al. (2009)

*Clustered regularly interspaced short palindromic repeats (CRISPR), recombinase polymerase amplification (RPA), African swine fever virus (ASFV), Recombinase polymerase amplification (RPA), Nucleic Acid Sequence Based Amplification (NASBA), Rolling circle amplification (RCA), enzyme-linked immunosorbent assay (ELISA), Single-molecule enzyme-linked immunosorbent assay (SIMOA).

ELISA and many more. The exploration of such attractive approaches is in the initial stage and thus these techniques would be employed to detect SARS-CoV-2 infections in local settings. Emerging diagnostic technologies that can play a key role in facilitating POC approaches should be further investigated to control the current SARS-CoV-2 and future outbreaks.

The diagnostic technologies currently in development can be classified into at least four phases (Fig. 3). Phase 1 consists of technologies in the proof-of-concept stage in research and development laboratories where researchers may use synthetic biomarker targets to confirm the concept. In phase 2, the newly developed diagnostic technologies are used to analyze a small number of patient samples in independent groups. Both phases 1 and 2 are normally conducted in academic laboratory settings, whereas phases 3 and 4 are conducted by private enterprises after commercial handover and the developed technology is subsequently made available for patients. Most alternative diagnostic technologies are still in the first "proof-of-concept" stage and only a few have passed the first two phases. The diagnostic technologies in phase 3 should be quickly adopted by private enterprises for POC and large-scale diagnosis to control the emergent SARS-CoV-2 outbreaks. The progression of phase 2 technologies into phase 3 (after clinical trials) will increase the availability of alternative diagnostic techniques for implementing rapid and POC approaches for detecting new SARS-CoV-2-infected patients. An accurate and robust point-of-care (POC) diagnostic infrastructure would also needs to be designed for achieving public health security, early diagnosis, disease surveillance, and ease of contact tracing (Majors et al., 2017; Nayak et al., 2017; Peeling and Mabey 2010). POC testing needs to be used timely for rapid diagnosis of infectious diseases particularly.

7. Public health surveillance and mobile-health

Monitoring an epidemic crisis requires constant surveillance, retroactive tracking, epidemiological database procurement, and regular

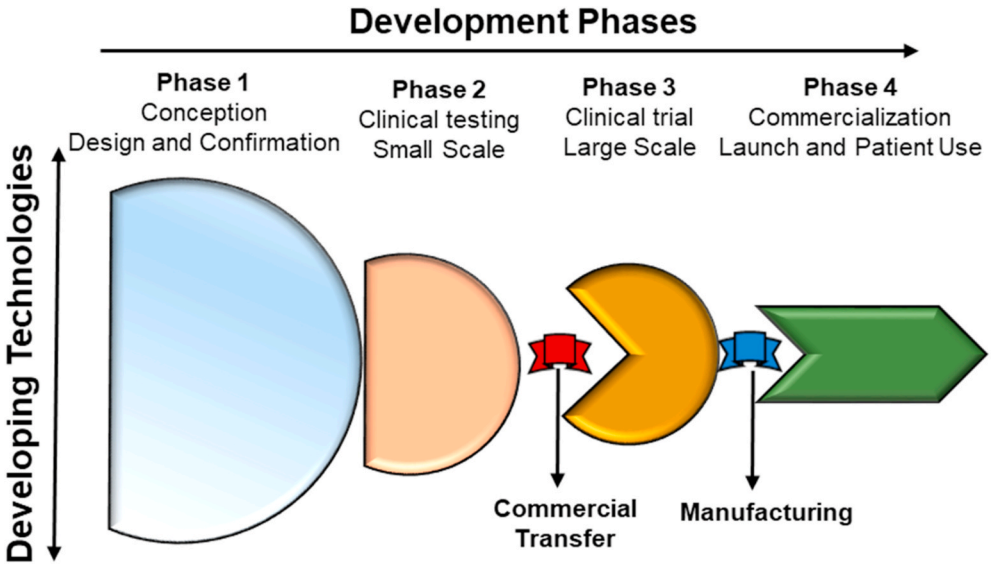

Fig. 3. Developmental phases for diagnostic tests. Academic laboratories are responsible for phases 1 and 2, while companies and private enterprises have to perform phases 3 and 4 after the commercial transfer occurs. Most emerging diagnostic tools are in the first proof-of-concept phase, and those in phase 3 will be rapidly adapted for the diagnosis of SARS-CoV-2. The progression of these technologies from phase 2 to phase 3 will increase the series of integral approaches for rapid and POC detection of SARS-CoV-2.

monitoring of patients. Healthcare providers, from local community health centers to treatment hospitals, must apply communication tools that can increase the speed and ease of surveillance to curb the spread of the infection. Smartphone networks can be leveraged for surveillance purposes as they provide benefits such as electronic data processing, a widespread network, and essential hardware to simplify electronic reporting, epidemiological database creation, and POC diagnosis (Fig. 4). Smartphone adoption and networks have increased exponentially worldwide, including in developing Asian and African countries. Smartphones are now widely accessible tools that can be used to coordinate surveillance and responses during existing global outbreaks like SARS-CoV-2 (Nayak et al., 2017; Wood et al., 2019).

The global spread of SARS-CoV-2 has been accelerated by poor communication and underreporting (Sun et al., 2020; Zhang et al., 2020c). One important example is in Iran, where 43 cases were initially confirmed on 23 February 2020; however, the fatality rate was about 19%, with 8 deaths and 3 other cases of Iranian citizens (2020b). This suggested that the number of infected patients in Iran was in the range of thousands according to transmission modeling. There were numerous uncertainties about the severity of the SARS-CoV-2 epidemic in Iran, and several field-level epidemiologic surveys and surveillance modules are currently being applied to produce more reliable estimates.

There have been noteworthy advances in the integration of diagnostic tools and smartphones. Smartphone accessories (e.g., digital camera, audio, and flashlights) can provide readout signals for diagnostic results as an alternative to local laboratory settings (Malekjahani et al., 2019). Smartphone devices can streamline the workflow of diagnosis and reporting by automating signal readout and database creation. For instance, smartphones connected to a microscope were tested recently for POC and were found to provide faster processing of blood-borne filarial parasite samples than the standard procedure (D'Ambrosio et al., 2015). Kanazawa et al. demonstrated the use of a smartphone application for thermography to detect hyper-inflammation and body temperature (Kanazawa et al. (2016)). This smartphone-based thermographic technology can also be used for the detection of common symptoms, comprising hyper-inflammation and fever caused by different infections and coronaviruses including SARS-CoV-2. Mudanyali et al. previously developed a rapid diagnostic test for malaria, tuberculosis, and HIV using a microscope attached to a smartphone that transferred the data to diagnostic test reader platforms and subsequently to a database app for spatiotemporal analysis and mapping of the results (Mudanyali et al., 2012a). Such devices are appropriate to address the underreported infections needed for rapid POC diagnostic tests at the local community level and at healthcare providers.

Furthermore, smartphones can be connected to existing diagnostic tools to provide real-time geospatial data that enable national and international healthcare agencies to implement organized strategies to control the pandemic. Smartphones can digitize the process of contact tracing and surveillance to provide wide-ranging and shareable data. The powerful portable processor with wireless connectivity and sensors

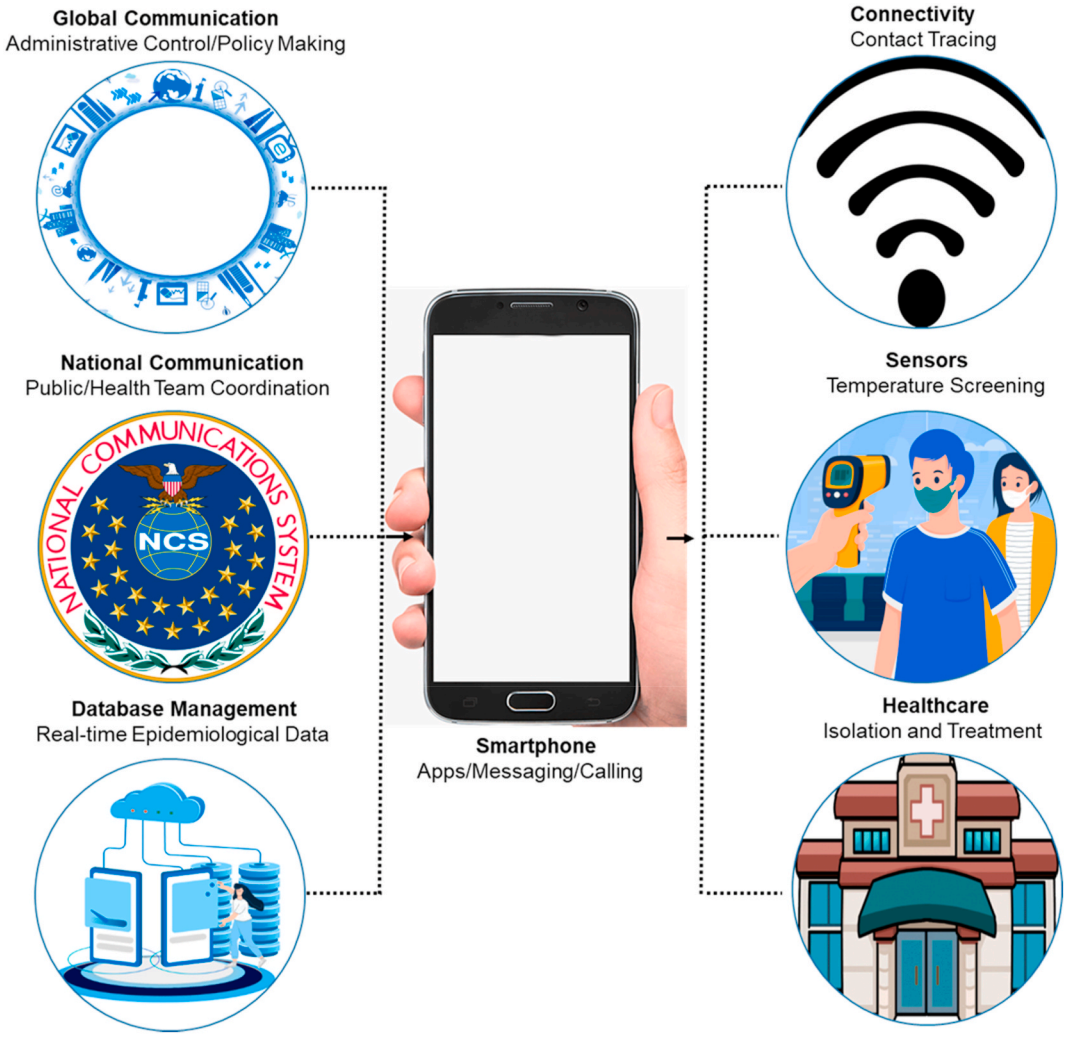

Fig. 4. Applications of smartphones in communication diagnostics and coordination with healthcare providers. Smartphones are useful for local, national, and global connectivity as well as policy-making based on data and experiences.

can enable scientists and healthcare providers to accurately generate and handle surveillance data (Perkel 2017). The capabilities and global adoption of smartphones and mobile-friendly technologies that are applicable in both resource-limited and resource-rich settings are growing exponentially (Oliver et al., 2020). Furthermore, the low cost of smartphones mitigates challenges such as the affordability barrier while offering POC detection and data processing facilities similar to those achieved with 'high-end' and more costly services. Several researchers have successfully used smartphones for geo-spatial data tracking for infectious diseases such as Ebola, HIV, and tuberculosis (Brangel et al., 2018; Danquah et al., 2019; Iribarren et al., 2016). In recent pandemics including MERS-CoV, Ebola SARS-CoV-1, and SARS-CoV-2, smartphones were successfully used for contact tracing and for extensive digital and real-world surveillance to identify and track individuals that came in contact with infected patients to curb further spread.

The development of smartphone-based diagnostic devices began with attached exterior accessories (D'Ambrosio et al., 2015; Laksanasopin et al., 2015), accessory-free diagnostic systems (Cho et al., 2015a), paper-based assays (Kong et al., 2019), serological POC tests (Brangel et al., 2018; Wang et al., 2011b), and tools that interface with smartphones (D'Ambrosio et al., 2015; Snodgrass et al., 2018). Real-time diagnosis of viral-induced reactive oxygen species in fresh sputum was reported by using an electrochemical system during the existing COVID-19 pandemic (Miripour et al., 2020). A detailed review of biosensor-based electrochemical pathogen detection was recently reported for emerging electrodes allow efficient transduction of selectively pathogen binding (Cesewski and Johnson 2020). These studies showed that such diagnostic platforms are capable of detecting signal outputs based on the principles of fluorescence, colorimetry, microscopy, and electrochemistry (Table 2) for clinically validated and approved methods.

Correspondence barriers may exist between suspected SARS-CoV-2 patients and healthcare providers due to patient hesitation or other personal reasons. Patients who are asymptomatic or show mild respirational symptoms may hesitate to visit overcrowded hospital centers, as they may face a higher risk of contracting SARS-CoV-2 at these locations. Telephone networks can facilitate direct communication between suspected patients and healthcare providers without the risk of infection to either party. Telemedicine was successfully used to manage suspected patients during the influenza pandemic in Switzerland in 2009 (Blozik (2017)). This approach led to more reporting of infected cases as compared to face-to-face consultations due to the lower risk.

Table 2
List of smartphone-based diagnostic applications for evaluating different types of diseases.

Readout Signal	Diseases	Biomarkers	Analytical sensitivity	Clinical specificity	Clinical sensitivity	POC Y/N	References
Colorimetric	SARS-CoV-2	RT-LAMP	10 copies	–	~97%	N	Yu et al. (2020a)
	Measles	Measles IgG	–	~97%	~96%	Y	Berg et al. (2015)
	Herpes simplex virus (HSV) −1/2	Mumps IgG	–	~95%	99%	Y	Berg et al. (2015)
	Mumps	HSV-1/2 IgG	–	~97, ~96%,	~98, ~99%	Y	Berg et al. (2015)
	Ebola	anti-IgG for SUDV GPl-649, EBOV GP1-649, and BDBV GP1-649	200 ng/mL	~98%	~97%	Y	Brangel et al. (2018)
	Ovarian cancer	HE4	20 ng/mL	~90%	~89.5%	N	Wang et al. (2011a)
	Zika	NS-1	0.05 ng/mL	–	–	Y	Rong et al. (2019)
	Urinary tract infection	*E. coli, Neisseria gonorrhoeae*	10 CFU/mL	–	–	N	Cho et al. (2015b)
	Human immunodeficiency virus (HIV)	p24	1 pg/mL	–	–	N	Loynachan et al. (2018)
	Dengue	Dengue viral DNA	5 nM	–	–	N	Choi et al. (2016)
	HIV, Tuberculosis, malaria	HIV 1/2 IgG, TB IgG, *P. falciparum*	–	–	–	N	Mudanyali et al. (2012b)
Fluorescence	Avian influenza	H5N1 nucleoprotein	8×10^5 PFU/mL	~99%	~96%	Y	Yeo et al. (2016)
	Ebola	EBOV glycoprotein	0.2 ng/mL	–	–	N	Hu et al. (2017)
	Thrombin	Thrombin	18 NIH units/mL	–	–	N	Petryayeva and Algar (2015)
	HIV	p24	17 pg/mL	–	–	N	Joh et al. (2017)
	Zika	RT-LAMP, Zika viral RNA, and whole virus	$3 \times x10^4$ PFU/mL (plaque-forming units)	–	–	Y	Ganguli et al. (2017)
	Cytomegalovirus	HMCV	1×10^3 PFU/mL	–	–	N	Wei et al. (2013)
	HSV-2	HSV-2 viral DNA	100 copies/pL	–	–	Y	Liao et al. (2016)
	HIV, dengue	anti-HIV1-p17, anti-NS1	100 pM	–	–	N	Arts et al. (2016)
	HIV, Hepatitis B (HBV)	HIV viral cDNA, HBV viral DNA	1×10^3 copies/mL	–	–	N	Ming et al. (2015)
	Zika, dengue, chikungunya	ZIKV, DENV, and CHIKV viral RNA	22 PFU/mL	–	–	N	Priye et al. (2017)
	E. coli	*E. coli* DNA	300 copies/pL	–	–	Y	Stedtfeld et al. (2012)
Electrochemical	Human papillomavirus	HPV viral DNA	50 amol	90–97%	83–92%	N	Ho et al. (2018)
	Sepsis	IL-3	22 pg/mL	~82%	~91%	Y	Min et al. (2018)
	Syphilis, HIV	treponemal syphilis antibody, HIV 1/2	2 pg/mL	~89%, ~91%	~77%, ~98%	Y	Laksanasopin et al. (2015)
	Cervical cancer	HPV viral DNA	10 amol	~92%	~95%		Im et al. (2015)
	HIV	p24, anti-p24	48 ng/mL	~97%	~98%		Turbé et al. (2017)
	HCV	Hepatitis C core antibody	12 pM	–	–		Aronoff-Spencer et al. (2016)
	Malaria	PfHRP2	20 ng/mL	–	–		Lillehoj et al. (2013)
Microscopic	Schistosomiasis	*S. hematobium* ova	–	~93%	~56%		Ephraim et al. (2015)
	Loa loa filariasis	L. loa microfilariae	3×10^4 mf/mL	~94%	~96%		D'Ambrosio et al. (2015)
	Giardiasis	*G. lamblia* cysts	1×10^6 cells/mL	–	–		Koydemir et al. (2015)
	Malaria, tuberculosis	*P. falciparum* smear, M.	–	–	–		Breslauer et al. (2009)
	S. aureus	*S. aureus* cells	50 cfu/mL (colony forming units)	–	–		Shrivastava et al. (2018)

If individuals that may be infected with SARS-CoV-2 visit test centers and receive positive test results, infected patients with mild symptoms are usually sent home and instructed to self-quarantine. Self-quarantine deters prompt communication between healthcare providers and infected patients, causing contrary anxiety problems for such patients, who lack access to the active monitoring provided to the admitted patients by the clinician. Smartphone apps can connect patients with mental health counselors to help them cope with anxiety resulting from isolation, fear of death during self-quarantine, and the epidemic (Liu et al., 2020d). SARS-CoV-2 patients can also report their daily symptoms and progress, simplifying remote monitoring by healthcare providers (Karimuribo et al., 2017). Smartphone app-based self-reporting also provides relevant information to healthcare providers with potential transmission pathways. For instance, a smartphone app was successfully used during the MERS outbreak in 2013 to monitor international travelers. With such an app, suspected patients could access the hygiene protocols and conduct self-reporting about animal exposure and the beginning of symptoms during their pilgrimage in Saudi Arabia as well as after returning to their respective home countries (Amani et al., 2015). Using smartphone apps developed for national and international service, public health providers and agencies can immediately make information available through public service, facilitating active reporting and prompt responses for the prevention of potential disease outbreaks.

Recent developments in mobile surveillance and POC testing have occurred with the emergence of mHealth, a term coined by the WHO to describe the use of mobile devices to assist public health and medical personnel practitioners (Ali et al., 2016). This has spurred the development of diagnostic tools and the creation of mobile devices connected to modern diagnostic tools. The development of real-time mHealth solutions can be used to address a range of problems and challenges in the fields of emerging disease surveillance, healthcare, and mental health counseling (Mackillop et al., 2014). The integration of diagnostics tools has impacted all areas of healthcare and public health concerns, including the control of infectious disease outbreaks.

Though precise contact tracing comes with considerable challenges, recent developments have demonstrated the potential benefits of mHealth in pandemic settings. Smartphone apps developed nationally and internationally have improved data collection, storage, and accurate policy implementation. However, the challenges of using smartphone apps in an epidemic and emergency include human rights violations and the lack of data security to ensure reliable privacy and confidentiality. Optimal use of mHealth apps has the potential to result in the creation of the largest health database for use in research, surveillance, and crisis management interventions (Pisani et al., 2016; Tresp et al., 2016). Clinic-based facilities, where health data from the diagnostic analysis are currently acquired and deposited in local laboratory organizations are relatively secure. However, data collected from separate mHealth apps may not be stored securely or easily accessible across numerous mHealth apps and health databases connected electronically (Chen et al., 2012).

The latest ethical studies have stressed the need for people to understand and consent to all aspects of how their personal data will be utilized (Rumbold et al., 2017). Integrated platforms to share data, standardize security, organize collected data, and improve mHealth app connectivity and services are presented in the developmental stage (Chen et al., 2012). However, though these mHealth platforms have potential, including rapid healthcare interoperability, a substantial collaboration between investors and regulation of the interfaces in mHealth services are still necessary before they can be implemented. The rapidly developing arena, however, requires both the improvement of regulatory policies that may encompass the wide range of medical-related smartphone apps currently in development.

8. Conclusions and future perspectives

In conclusion, development of SARS-CoV-2 diagnostic methods from phase 1 to phase 4 has been expedited, allowing investigators to contribute to the conception, design, and validation of these technologies. The development of diagnostic technologies usually requires several years for design optimization and the completion of all required trials, but scientists are now playing a key role in the development process to enable prompt diagnosis and curb the spread of SARS-CoV-2. The morphology of the SARS-CoV-2 virus was identified using TEM, complete genome sequencing was performed to confirm the biological characteristics, and the sequencing data were successfully used to design PCR probes and primers. Compared to SARS-CoV, which took several months to identify, the SARS-CoV-2 virus was identified promptly, enabling the rapid development of sophisticated diagnostic kits and their provision to healthcare providers. Several techniques were developed within 3–4 weeks to identify SARS-CoV-2 with high specificity and sensitivity. The rapid sequencing of SARS-CoV-2 and identification of probes and primers has enabled scientific authorities to rapidly develop nucleic acid-based test kits. These strategies successfully provided the first line of defense against the outbreak; however, it remains a significant pandemic worldwide. Various strategies are being developed to establish serological tests, SARS-CoV-2 antigen S protein and N protein testing, microfluidic chemiluminescent ELISA system, SARS-CoV-2 IgG and IgM testing using well-established technologies, since those kits would be easier to handle, can be used for POC services, and serve as a complement to nucleic acid-based test kits developed for the diagnosis of SARS-CoV-2-suspected patients.

A detailed review of recent literature and future perspectives on potential biosensors developed for point-of-care, high-sensitivity, and rapid detection of SARS-CoV-2 infections (Ji et al., 2020). Further developments in nucleic acid-based diagnostic kits for the diagnosis of SARS-CoV-2 have occurred in recent months. New and more efficient diagnostic technologies are in development to provide cost-effective tools that are appropriate for POC tests and can be applied for large populations. A recently published review also provided a comprehensive report on molecular nucleic acid assays and immunological and serological tests for the diagnosis of SARS-CoV-2 infection (Ravi et al., 2020). However, other types of assay based on nucleic acids including hybridization microarray assay, isothermal amplification, metagenomics sequencing, and CRISPR are under development and some have been approved for general use (Huang et al., 2020c). Studies investigating the success of POC test kits and multiplex assays for the diagnosis of both suspected patients and recovered asymptomatic patients are warranted for accurate estimation of the SARS-CoV-2 epidemiological data. Emerging diagnostic technologies currently in phases 2 and 3 include isothermal amplification, microfluidic technologies, barcoding, and protein-based assays. These technologies should be quickly made available to healthcare providers for the implementation of POC systems that can be employed to curb the outbreak. The integration of smartphones and diagnostic tools may provide a greater opportunity to improve diagnostic practices, surveillance, contact tracing, and mental health counseling services.

Significant progress is anticipated in the development of novel diagnostic tests despite various challenges. Tireless efforts from the global research community coupled with the sharing of technologies have facilitated the development of new diagnostic assays and success in the vision of worldwide test kit delivery. To promote the development of faster, sensitive, and accurate diagnostic tool kits, a number of research organizations are supporting research and development efforts by submitting test tools for evaluation or by providing needed funds for greater collaboration. Due to several initiatives and technological advancements, including cooperative scientific developments, diagnostic technologies for SARS-CoV-2 will likely continue to improve and thrive in a near future. In the midst of infectious disease, diagnostic devices with connectivity provide an opportunity to develop disruptive diagnostic

technologies with the potential to meet the requirements of healthcare systems. These developments should increase direct access to diagnosis, contact testing, and treatment options for SARS-CoV-2-infected patients while improving epidemic assessment, infection surveillance, and the control of public health with great precision. In addition to these sophisticated technologies, the potential public-health benefit is a prerequisite, whichever through the personal, community, and social responsibilities have to be strictly followed in an outbreak like situation. Controlling pandemics through collective-responsibility or 'social science' initiatives has huge significance and also through high level alarming messaging since individuals and communities have 3G or 4G access of digital services, and finally with the assistance of public health providers and clinical systems providing early detection, clinical care and spread control.

To take advantage of recent developments, advances in early diagnostics with an accurate understanding of the outbreak context are necessary and participants may utilize scientific resources readily associated with smartphone systems. Systems within resource-rich settings may differ from those in resource-limited surroundings; therefore, different technical ecosystems may be needed to address specific challenges. The demand for diagnostic tools in resource-rich locations may drive development and commercial production in future, but the potential of these tools to transform healthcare in poorer countries after appropriately tailoring them to local needs should also be investigated. To achieve this, large investments in research and development are needed for the development of appropriate devices, tools, and systems that are applicable in all socio-economic settings to address the existing divide in digital services. To achieve this, stringent regulations and governance policies for mHealth disruptive technologies and devices as well as their deployment must be resolved by simultaneously addressing the existing challenges of diagnosis, contact testing, disease surveillance, data security, digital access, and autonomy for clinical authorities. It is evident that future diagnostic tools and kits are likely to be connected and digital, accelerating transformation in the healthcare system. Thus, scientists and policymakers have an exciting opportunity and a challenging task to promptly deploy these tools to transform and improve existing healthcare systems. It is also important to note that scientific reports on SARS-CoV-2 are evolving rapidly; therefore, some of the information presented in this review may change as new studies become available.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

The authors express appreciation to the Dongguk University-Seoul for Research Fund (RF: 2020-2022). The authors extend their appreciation to the Deanship of Scientific Research at King Saud University for funding this work through research group No (RG-1441-313).

References

Abbasi, J., 2020. JAMA 323 (19), 1881–1883.
Abudayyeh, O.O., Gootenberg, J.S., Essletzbichler, P., Han, S., Joung, J., Belanto, J.J., Verdine, V., Cox, D.B.T., Kellner, M.J., Regev, A., Lander, E.S., Voytas, D.F., Ting, A. Y., Zhang, F., 2017. Nature 550 (7675), 280–284.
Agrati, C., D'Offizi, G., Narciso, P., Abrignani, S., Ippolito, G., Colizzi, V., Poccia, F., 2001. Mol. Med. 7 (1), 11–19.
Ahmed, S.F., Quadeer, A.A., McKay, M.R., 2020. Viruses 12 (3), 254.
Ai, T., Yang, Z., Hou, H., Zhan, C., Chen, C., Lv, W., Tao, Q., Sun, Z., Xia, L., 2020. Radiology 296 (2), E32–E40.
Ali, E.E., Chew, L., Yap, K.Y.-L., 2016. BMJ Innovations 2 (1), 33–40.
Amani, S.A., Nasser, F.B., Mohamed, T., Harold, W.W., Kerrie, E.W., Anita, E.H., Robert, B., Harunor, R., 2015. J. Epidemiol. Glob. Health 6 (3), 147–155.
Amen, et al., 2020. Nat. Biotechnol. 38 (7), 791–797.
Amer, H.M., Abd El Wahed, A., Shalaby, M.A., Almajhdi, F.N., Hufert, F.T., Weidmann, M., 2013. J. Virol. Methods 193 (2), 337–340.
Anasir, M.I., Poh, C.L., 2019. Front. Microbiol. 10 (738).
Andersen, K.G., Rambaut, A., Lipkin, W.I., Holmes, E.C., Garry, R.F., 2020. Nat. Med. 26 (4), 450–452.
Aronoff-Spencer, E., Venkatesh, A.G., Sun, A., Brickner, H., Looney, D., Hall, D.A., 2016. Biosens. Bioelectron. 86, 690–696.
Arts, R., den Hartog, I., Zijlema, S.E., Thijssen, V., van der Beelen, S.H., Merkx, M., 2016. Anal. Chem. 88 (8), 4525–4532.
Atkinson, B., Petersen, E., 2020. Lancet 395 (10233), 1339–1340.
Aytur, T., Foley, J., Anwar, M., Boser, B., Harris, E., Beatty, P.R., 2006. J. Immunol. Methods 314 (1), 21–29.
Bagdonaite, I., Wandall, H.H., 2018. Glycobiology 28 (7), 443–467.
Belouzard, S., Chu, V.C., Whittaker, G.R., 2009. Proc. Natl. Acad. Sci. U.S.A. 106 (14), 5871–5876.
Berg, B., Cortazar, B., Tseng, D., Ozkan, H., Feng, S., Wei, Q., Chan, R.Y.-L., Burbano, J., Farooqui, Q., Lewinski, M., Di Carlo, D., Garner, O.B., Ozcan, A., 2015. ACS Nano 9 (8), 7857–7866.
Bernheim, A., Mei, X., Huang, M., Yang, Y., Fayad, Z.A., Zhang, N., Diao, K., Lin, B., Zhu, X., Li, K., Li, S., Shan, H., Jacobi, A., Chung, M., 2020. Radiology 295 (3), 200463.
Bicart-See, A., Rottman, M., Cartwright, M., Seiler, B., Gamini, N., Rodas, M., Penary, M., Giordano, G., Oswald, E., Super, M., Ingber, D.E., 2016. PloS One 11 (6), e0156287.
Biron, C.A., Byron, K.S., Sullivan, J.L., 1989. N. Engl. J. Med. 320 (26), 1731–1735.
Blanco-Melo, D., Nilsson-Payant, B.E., Liu, W.C., Uhl, S., Hoagland, D., Møller, R., Jordan, T.X., Oishi, K., Panis, M., Sachs, D., Wang, T.T., Schwartz, R.E., Lim, J.K., Albrecht, R.A., tenOever, B.R., 2020. Cell 181 (5), 1036–1045 e1039.
Bloch, et al., 2020. J. Clin. Invest. 130 (6), 2757–2765.
Blozik, et al., 2017. Sci. Transl. Med. 9 (409) eaan1589.
Brangel, P., Sobarzo, A., Parolo, C., Miller, B.S., Howes, P.D., Gelkop, S., Lutwama, J.J., Dye, J.M., McKendry, R.A., Lobel, L., Stevens, M.M., 2018. ACS Nano 12 (1), 63–73.
Breslauer, D.N., Maamari, R.N., Switz, N.A., Lam, W.A., Fletcher, D.A., 2009. PloS One 4 (7), e6320.
Broughton, J.P., Deng, X., Yu, G., Fasching, C.L., Servellita, V., Singh, J., Miao, X., Streithorst, J.A., Granados, A., Sotomayor-Gonzalez, A., Zorn, K., Gopez, A., Hsu, E., Gu, W., Miller, S., Pan, C.-Y., Guevara, H., Wadford, D.A., Chen, J.S., Chiu, C.Y., 2020. Nat. Biotechnol. 38 (7), 870–874.
Brouwer, et al., 2020. Science 369 (6504), 643–650.
Bui, H., Díaz, S.A., Fontana, J., Chiriboga, M., Veneziano, R., Medintz, I.L., 2019. Adv. Opt. Mater. 7 (18), 1900562.
Cai, et al., 2020. J. Infect. Dis. 222 (2), 189–193.
Cai, X., Sushkov, A.B., Suess, R.J., Jadidi, M.M., Jenkins, G.S., Nyakiti, L.O., Myers-Ward, R.L., Li, S., Yan, J., Gaskill, D.K., Murphy, T.E., Drew, H.D., Fuhrer, M.S., 2014. Nat. Nanotechnol. 9 (10), 814–819.
Carsana, L., Sonzogni, A., Nasr, A., Rossi, R.S., Pellegrinelli, A., Zerbi, P., Rech, R., Colombo, R., Antinori, S., Corbellino, M., Galli, M., Catena, E., Tosoni, A., Gianatti, A., Nebuloni, M., 2020. Lancet Infect. Dis. 20 (10), 1135–1140.
Carter, L.J., Garner, L.V., Smoot, J.W., Li, Y., Zhou, Q., Saveson, C.J., Sasso, J.M., Gregg, A.C., Soares, D.J., Beskid, T.R., Jervey, S.R., Liu, C., 2020. ACS Cent. Sci. 6 (5), 591–605.
Casrouge, A., Bisiaux, A., Stephen, L., Schmolz, M., Mapes, J., Pfister, C., Pol, S., Mallet, V., Albert, M.L., 2012. Clin. Exp. Immunol. 167 (1), 137–148.
Cesewski, E., Johnson, B.N., 2020. Biosens. Bioelectron. 159, 112214.
Chan, C.-M., Woo, P.C.Y., Lau, S.K.P., Tse, H., Chen, H.-L., Li, F., Zheng, B.-J., Chen, L., Huang, J.-D., Yuen, K.-Y., 2008. Exp. Biol. Med. 233 (12), 1527–1536.
Chauhan, G., Madou, M.J., Kalra, S., Chopra, V., Ghosh, D., Martinez-Chapa, S.O., 2020. ACS Nano 14 (7), 7760–7782.
Che, X.-y., Qiu, L.-w., Pan, Y.-x., Wen, K., Hao, W., Zhang, L.-y., Wang, Y.-d., Liao, Z.-y., Hua, X., Cheng, V.C.C., Yuen, K.-y., 2004. J. Clin. Microbiol. 42 (6), 2629–2635.
Chen, Z., John Wherry, E., 2020. Nat. Rev. Immunol. 20 (9), 529–536.
Chen, C., Haddad, D., Selsky, J., Hoffman, J.E., Kravitz, R.L., Estrin, D.E., Sim, I., 2012. J. Med. Internet Res. 14 (4), e112.
Chen, J.S., Ma, E., Harrington, L.B., Da Costa, M., Tian, X., Palefsky, J.M., Doudna, J.A., 2018. Science 360 (6387), 436–439.
Chen, W.-H., Hotez, P.J., Bottazzi, M.E., 2020a. Hum. Vaccines Immunother. 16 (6), 1239–1242.
Chen, Y., Guo, Y., Pan, Y., Zhao, Z.J., 2020b. Biochem. Biophys. Res. Commun. 525 (1), 135–140.
Chen, Y., Wang, J., Liu, C., Su, L., Zhang, D., Fan, J., Yang, Y., Xiao, M., Xie, J., Xu, Y., Li, Y., Zhang, S., 2020c. Mol. Med. 26 (1), 97.
Chen, Z., Wu, Q., Chen, J., Ni, X., Dai, J., 2020d. Virol. Sin. 35 (3), 351–354.
Chi, X., Yan, R., Zhang, J., Zhang, G., Zhang, Y., Hao, M., Zhang, Z., Fan, P., Dong, Y., Yang, Y., Chen, Z., Guo, Y., Zhang, J., Li, Y., Song, X., Chen, Y., Xia, L., Fu, L., Hou, L., Xu, J., Yu, C., Li, J., Zhou, Q., Chen, W., 2020. Science 369 (6504), 650–655.
Chin, et al., 2011. Nat. Med. 17 (8), 1015–1019.
Cho, S., Park, T.S., Nahapetian, T.G., Yoon, J.-Y., 2015a. Biosens. Bioelectron. 74, 601–611.
Cho, S., Park, T.S., Nahapetian, T.G., Yoon, J.Y., 2015b. Biosens. Bioelectron. 74, 601–611.
Choi, J.R., Hu, J., Feng, S., Wan Abas, W.A.B., Pingguan-Murphy, B., Xu, F., 2016. Biosens. Bioelectron. 79, 98–107.
Chu, D.K.W., Pan, Y., Cheng, S.M.S., Hui, K.P.Y., Krishnan, P., Liu, Y., Ng, D.Y.M., Wan, C.K.C., Yang, P., Wang, Q., Peiris, M., Poon, L.L.M., 2020. Clin. Chem. 66 (4), 549–555.
Clery, D., 2002. Science 297 (5582), 761–763.

Coperchini, F., Chiovato, L., Croce, L., Magri, F., Rotondi, M., 2020. Cytokine Growth Factor Rev. 53, 25–32.
Corman, et al., 2020. Euro Surveill. 25 (3), 2000045.
Correia, et al., 2014. Nature 507 (7491), 201–206.
Craw, P., Balachandran, W., 2012. Lab Chip 12 (14), 2469–2486.
Crivellato, E., Travan, L., Ribatti, D., 2010. Int. Arch. Allergy Immunol. 151 (2), 89–97.
Cui, J., Li, F., Shi, Z.-L., 2019. Nat. Rev. Microbiol. 17 (3), 181–192.
Cunha, B.A., Pherez, F.M., Schoch, P., 2009. Clin. Infect. Dis. 49 (9), 1454–1456.
Damas, J., Hughes, G.M., Keough, K.C., Painter, C.A., Persky, N.S., Corbo, M., Hiller, M., Koepfli, K.-P., Pfenning, A.R., Zhao, H., Genereux, D.P., Swofford, R., Pollard, K.S., Ryder, O.A., Nweeia, M.T., Lindblad-Toh, K., Teeling, E.C., Karlsson, E.K., Lewin, H. A., 2020. Proc. Natl. Acad. Sci. U. S. A, 202010146.
Danquah, L.O., Hasham, N., MacFarlane, M., Conteh, F.E., Momoh, F., Tedesco, A.A., Jambai, A., Ross, D.A., Weiss, H.A., 2019. BMC Infect. Dis. 19 (1), 810.
De, S., Higgins, T.M., Lyons, P.E., Doherty, E.M., Nirmalraj, P.N., Blau, W.J., Boland, J.J., Coleman, J.N., 2009. ACS Nano 3 (7), 1767–1774.
Deng, X., Li, L., Enomoto, M., Kawano, Y., 2019. Sci. Rep. 9 (1), 3498.
Denison, M.R., Graham, R.L., Donaldson, E.F., Eckerle, L.D., Baric, R.S., 2011. RNA Biol. 8 (2), 270–279.
Dhama, K., Khan, S., Tiwari, R., Sircar, S., Bhat, S., Malik, Y.S., Singh, K.P., Chaicumpa, W., Bonilla-Aldana, D.K., Rodriguez-Morales, A.J., 2020. Clin. Microbiol. Rev. 33 (4) e00028-00020.
Dharavath, B., Yadav, N., Desai, S., Sunder, R., Mishra, R., Ketkar, M., Bhanshe, P., Gupta, A., Redhu, A.K., Patkar, N., Dutt, S., Gupta, S., Dutt, A., 2020. Heliyon 6 (7), e04405.
Dong, Y., Dai, T., Wei, Y., Zhang, L., Zheng, M., Zhou, F., 2020. Signal Transduction and Targeted Therapy 5 (1), 237.
Dowdy, et al., 2020. Proc. Natl. Acad. Sci. U.S.A. 117 (17), 9490–9496.
Duan, L., Zheng, Q., Zhang, H., Niu, Y., Lou, Y., Wang, H., 2020. Front. Immunol. 11 (2593).
Dunning, J., Blankley, S., Hoang, L.T., Cox, M., Graham, C.M., James, P.L., Bloom, C.I., Chaussabel, D., Banchereau, J., Brett, S.J., Moffatt, M.F., O'Garra, A., Openshaw, P. J.M., 2018. Nat. Immunol. 19 (6), 625–635.
D'Ambrosio, M.V., Bakalar, M., Bennuru, S., Reber, C., Skandarajah, A., Nilsson, L., Switz, N., Kamgno, J., Pion, S., Boussinesq, M., Nutman, T.B., Fletcher, D.A., 2015. Sci. Transl. Med. 7 (286), 286re284-286re284.
El Zowalaty, M.E., Järhult, J.D., 2020a. One Health (Amsterdam, Netherlands), vol. 9, 100124-100124.
El Zowalaty, M.E., Järhult, J.D., 2020b. One Health 9, 100124.
Ephraim, R.K., Duah, E., Cybulski, J.S., Prakash, M., D'Ambrosio, M.V., Fletcher, D.A., Keiser, J., Andrews, J.R., Bogoch II, 2015. Am. J. Trop. Med. Hyg. 92 (6), 1253–1256.
Fang, Y., Zhang, H., Xie, J., Lin, M., Ying, L., Pang, P., Ji, W., 2020. Radiology 200432, 0 (0).
Feng, W., Newbigging, A.M., Le, C., Pang, B., Peng, H., Cao, Y., Wu, J., Abbas, G., Song, J., Wang, D.-B., Cui, M., Tao, J., Tyrrell, D.L., Zhang, X.-E., Zhang, H., Le, X.C., 2020. Anal. Chem. 92 (15), 10196–10209.
Ferreira da Mota, N.V., Brunialti, M.K.C., Santos, S.S., Machado, F.R., Assuncao, M., Azevedo, L.C.P., Salomao, R., 2018. Shock 50 (3), 293–300.
Ferretti, L., Wymant, C., Kendall, M., Zhao, L., Nurtay, A., Abeler-Dörner, L., Parker, M., Bonsall, D., Fraser, C., 2020. Science 368 (6491) eabb6936.
Follis, K.E., York, J., Nunberg, J.H., 2006. Virology 350 (2), 358–369.
Fomsgaard, A.S., Rosenstierne, M.W., 2020. Euro Surveill. 25 (14), 2000398.
Foudeh, A.M., Fatanat Didar, T., Veres, T., Tabrizian, M., 2012. Lab Chip 12 (18), 3249–3266.
Freeman, W.M., Walker, S.J., Vrana, K.E., 1999. Biotechniques 26 (1), 112–125.
Ganguli, A., Ornob, A., Yu, H., Damhorst, G.L., Chen, W., Sun, F., Bhuiya, A., Cunningham, B.T., Bashir, R., 2017. Biomed. Microdevices 19 (4), 73.
Ganguli, A., Mostafa, A., Berger, J., Aydin, M.Y., Sun, F., Ramirez, S.A.S.d., Valera, E., Cunningham, B.T., King, W.P., Bashir, R., 2020. Proc. Natl. Acad. Sci. Unit. States Am. 117 (37), 22727–22735.
GeurtsvanKessel, C.H., Okba, N.M.A., Igloi, Z., Bogers, S., Embregts, C.W.E., Laksono, B. M., Leijten, L., Rokx, C., Rijnders, B., Rahamat-Langendoen, J., van den Akker, J.P. C., van Kampen, J.J.A., van der Eijk, A.A., van Binnendijk, R.S., Haagmans, B., Koopmans, M., 2020. Nat. Commun. 11 (1), 3436.
Ghnewa, Y.G., Fish, M., Jennings, A., Carter, M.J., Shankar-Hari, M., 2020. Med. Klin. Intensivmed. Notfallmed. 115 (Suppl. 1), 10–14.
Goo, J., Jeong, Y., Park, Y.-S., Yang, E., Jung, D.-I., Rho, S., Park, U., Sung, H., Park, P.-G., Choi, J.-a., Seo, S.H., Cho, N.H., Lee, H., Lee, J.M., Kim, J.-O., Song, M., 2020. Virus Res. 278, 197863.
Gootenberg, J.S., Abudayyeh, O.O., Lee, J.W., Essletzbichler, P., Dy, A.J., Joung, J., Verdine, V., Donghia, N., Daringer, N.M., Freije, C.A., Myhrvold, C., Bhattacharyya, R.P., Livny, J., Regev, A., Koonin, E.V., Hung, D.T., Sabeti, P.C., Collins, J.J., Zhang, F., 2017. Science 356 (6336), 438–442.
Grifoni, A., Sidney, J., Zhang, Y., Scheuermann, R.H., Peters, B., Sette, A., 2020a. Cell Host Microbe 27 (4), 671–680 e672.
Grifoni, et al., 2020b. Cell 181 (7), 1489–1501 e1415.
Guan, et al., 2020. N. Engl. J. Med. 382 (18), 1708–1720.
Guo, et al., 2020a. Nat. Commun. 11 (1), 3924.
Guo, L., Sun, X., Wang, X., Liang, C., Jiang, H., Gao, Q., Dai, M., Qu, B., Fang, S., Mao, Y., Chen, Y., Feng, G., Gu, Q., Wang, R.R., Zhou, Q., Li, W., 2020b. Cell Discovery 6 (1), 34.
Han, K.N., Li, C.A., Seong, G.H., 2013. Annu. Rev. Anal. Chem. 6 (1), 119–141.
Harmon, et al., 2020. Nat. Commun. 11 (1), 4080.
Harpaz, D., Eltzov, E., Ng, T.S.E., Marks, R.S., Tok, A.I.Y., 2020. Diagnostics 10 (1), 28.
Ho, N.R.Y., Lim, G.S., Sundah, N.R., Lim, D., Loh, T.P., Shao, H., 2018. Nat. Commun. 9 (1), 3238.
Hoffmann, M., Kleine-Weber, H., Schroeder, S., Krüger, N., Herrler, T., Erichsen, S., Schiergens, T.S., Herrler, G., Wu, N.-H., Nitsche, A., Müller, M.A., Drosten, C., Pöhlmann, S., 2020. Cell 181 (2), 271–280 e278.
Hou, Y., Peng, C., Yu, M., Li, Y., Han, Z., Li, F., Wang, L.-F., Shi, Z., 2010. Arch. Virol. 155 (10), 1563–1569.
Hu, J., Jiang, Y.-Z., Wu, L.-L., Wu, Z., Bi, Y., Wong, G., Qiu, X., Chen, J., Pang, D.-W., Zhang, Z.-L., 2017. Anal. Chem. 89 (24), 13105–13111.
Huang, K.J., Su, I.J., Theron, M., Wu, Y.C., Lai, S.K., Liu, C.C., Lei, H.Y., 2005. J. Med. Virol. 75 (2), 185–194.
Huang, P., Wang, H., Cao, Z., Jin, H., Chi, H., Zhao, J., Yu, B., Yan, F., Hu, X., Wu, F., Jiao, C., Hou, P., Xu, S., Zhao, Y., Feng, N., Wang, J., Sun, W., Wang, T., Gao, Y., Yang, S., Xia, X., 2018. Front. Microbiol. 9 (1101).
Huang, A.T., Garcia-Carreras, B., Hitchings, M.D.T., Yang, B., Katzelnick, L.C., Rattigan, S.M., Borgert, B.A., Moreno, C.A., Solomon, B.D., Trimmer-Smith, L., Etienne, V., Rodriguez-Barraquer, I., Lessler, J., Salje, H., Burke, D.S., Wesolowski, A., Cummings, D.A.T., 2020a. Nat. Commun. 11 (1), 4704.
Huang, C., Wang, Y., Li, X., Ren, L., Zhao, J., Hu, Y., Zhang, L., Fan, G., Xu, J., Gu, X., Cheng, Z., Yu, T., Xia, J., Wei, Y., Wu, W., Xie, X., Yin, W., Li, H., Liu, M., Xiao, Y., Gao, H., Guo, L., Xie, J., Wang, G., Jiang, R., Gao, Z., Jin, Q., Wang, J., Cao, B., 2020b. Lancet 395 (10223), 497–506.
Huang, Z., Tian, D., Liu, Y., Lin, Z., Lyon, C.J., Lai, W., Fusco, D., Drouin, A., Yin, X., Hu, T., Ning, B., 2020c. Biosens. Bioelectron. 164, 112316.
Iles, R.K., Zmuidinaite, R., Iles, J.K., Carnell, G., Sampson, A., Heeney, J.L., 2020a. Med. Rxiv. 2020.2008.2022.20176669.
Iles, R.K., Zmuidinaite, R., Iles, J.K., Carnell, G., Sampson, A., Heeney, J.L., 2020b. Diagnostics 10 (10), 746.
Im, H., Castro, C.M., Shao, H., Liong, M., Song, J., Pathania, D., Fexon, L., Min, C., Avila-Wallace, M., Zurkiya, O., Rho, J., Magaoay, B., Tambouret, R.H., Pivovarov, M., Weissleder, R., Lee, H., 2015. Proc. Natl. Acad. Sci. Unit. States Am. 112 (18), 5613–5618.
Imai, M., Ninomiya, A., Minekawa, H., Notomi, T., Ishizaki, T., Van Tu, P., Tien, N.T.K., Tashiro, M., Odagiri, T., 2007. J. Virol Methods 141 (2), 173–180.
Inui, S., Fujikawa, A., Jitsu, M., Kunishima, N., Watanabe, S., Suzuki, Y., Umeda, S., Uwabe, Y., 2020. Radiology: Cardiothoracic Imaging 2 (2), e200110.
Iribarren, S.J., Schnall, R., Stone, P.W., Carballo-Diéguez, A., 2016. JMIR mHealth uHealth 4 (2) e25.
Jain, J., Gaur, S., Chaudhary, Y., Kaul, R., 2020. VirusDisease 31 (2), 75–79.
Ji, T., Liu, Z., Wang, G., Guo, X., Akbar khan, S., Lai, C., Chen, H., Huang, S., Xia, S., Chen, B., Jia, H., Chen, Y., Zhou, Q., 2020. Biosens. Bioelectron. 166, 112455.
Jiang, S., Hillyer, C., Du, L., 2020. Trends Immunol. 41 (5), 355–359.
Jiao, J., Duan, C., Xue, L., Liu, Y., Sun, W., Xiang, Y., 2020. Biosens. Bioelectron. 167, 112479.
Jin, C., Chen, W., Cao, Y., Xu, Z., Tan, Z., Zhang, X., Deng, L., Zheng, C., Zhou, J., Shi, H., Feng, J., 2020. Nat. Commun. 11 (1), 5088.
Joh, D.Y., Hucknall, A.M., Wei, Q., Mason, K.A., Lund, M.L., Fontes, C.M., Hill, R.T., Blair, R., Zimmers, Z., Achar, R.K., Tseng, D., Gordan, R., Freemark, M., Ozcan, A., Chilkoti, A., 2017. Proc. Natl. Acad. Sci. Unit. States Am. 114 (34), E7054–E7062.
Ju, B., Zhang, Q., Ge, J., Wang, R., Sun, J., Ge, X., Yu, J., Shan, S., Zhou, B., Song, S., Tang, X., Yu, J., Lan, J., Yuan, J., Wang, H., Zhao, J., Zhang, S., Wang, Y., Shi, X., Liu, L., Zhao, J., Wang, X., Zhang, Z., Zhang, L., 2020. Nature 584 (7819), 115–119.
Kageyama, T., Kojima, S., Shinohara, M., Uchida, K., Fukushi, S., Hoshino, F.B., Takeda, N., Katayama, K., 2003. J. Clin. Microbiol. 41 (4), 1548–1557.
Kammila, S., Das, D., Bhatnagar, P.K., Sunwoo, H.H., Zayas-Zamora, G., King, M., Suresh, M.R., 2008. J. Virol Methods 152 (1), 77–84.
Kanazawa, R., Nakagami, R., Goto, R., Noguchi, H., Oe, R., Miyagaki, P., Hayashi, P., Sasaki, W., Sanada, R., 2016. J. Wound Care 25 (4), 177–182.
Karimuribo, E.D., Mutagahywa, E., Sindato, C., Mboera, L., Mwabukusi, M., Kariuki Njenga, M., Teesdale, S., Olsen, J., Rweyemamu, M., 2017. JMIR Public Health Surveill 3 (4), e94.
Kellner, M.J., Koob, J.G., Gootenberg, J.S., Abudayyeh, O.O., Zhang, F., 2019. Nat. Protoc. 14 (10), 2986–3012.
Kim, J., Maeng, I., Jung, J., Song, H., Son, J.-H., Kim, K., Lee, J., Kim, C.-H., Chae, G., Jun, M., Hwang, Y., Lee, S.J., Myoung, J.-M., Choi, H., 2013. Appl. Phys. Lett. 102 (1), 011109.
Kim, J., Biondi, M.J., Feld, J.J., Chan, W.C.W., 2016. ACS Nano 10 (4), 4742–4753.
Kim, J.-M., Chung, Y.-S., Jo, H.J., Lee, N.-J., Kim, M.S., Woo, S.H., Park, S., Kim, J.W., Kim, H.M., Han, M.-G., 2020. Osong Public Health Res Perspect 11 (1), 3–7.
Kirchdoerfer, R.N., Wang, N., Pallesen, J., Wrapp, D., Turner, H.L., Cottrell, C.A., Corbett, K.S., Graham, B.S., McLellan, J.S., Ward, A.B., 2018. Sci. Rep. 8 (1), 15701.
Kobayashi, Y., Mitsudomi, T., 2013. Transl. Lung Cancer Res. 2 (5), 354–363.
Kong, T., You, J.B., Zhang, B., Nguyen, B., Tarlan, F., Jarvi, K., Sinton, D., 2019. Lab Chip 19 (11), 1991–1999.
Koydemir, H.C., Gorocs, Z., Tseng, D., Cortazar, B., Feng, S., Chan, R.Y.L., Burbano, J., McLeod, E., Ozcan, A., 2015. Lab Chip 15 (5), 1284–1293.
Krafcikova, P., Silhan, J., Nencka, R., Boura, E., 2020. Nat. Commun. 11 (1), 3717.
Kubina, R., Dziedzic, A., 2020. Diagnostics (Basel) 10 (6), 434.
Kujawski, et al., 2020. Nat. Med. 26 (6), 861–868.
Kulesa, C., 2011. IEEE Transactions on Terahertz Science and Technology 1 (1), 232–240.
Kuri-Cervantes, et al., 2020. Sci Immunol 5 (49).
Kwong, P.D., 2017. Cold Spring Harb Perspect Biol 9 (11) a029470.
La Marca, A., Capuzzo, M., Paglia, T., Roli, L., Trenti, T., Nelson, S.M., 2020. Reprod. Biomed. Online 41 (3), 483–499.

Lafarge, X., Merville, P., Cazin, M.C., Bergé, F., Potaux, L., Moreau, J.F., Déchanet-Merville, J., 2001. J. Infect. Dis. 184 (5), 533–541.
Laing, et al., 2020. Nat. Med. 26 (10), 1623–1635.
Laksanasopin, T., Guo, T.W., Nayak, S., Sridhara, A.A., Xie, S., Olowookere, O.O., Cadinu, P., Meng, F., Chee, N.H., Kim, J., Chin, C.D., Munyazesa, E., Mugwaneza, P., Rai, A.J., Mugisha, V., Castro, A.R., Steinmiller, D., Linder, V., Justman, J.E., Nsanzimana, S., Sia, S.K., 2015. Sci. Transl. Med. 7 (273), 273re271-273re271.
Lan, J., Ge, J., Yu, J., Shan, S., Zhou, H., Fan, S., Zhang, Q., Shi, X., Wang, Q., Zhang, L., Wang, X., 2020. Nature 581 (7807), 215–220.
Lee, E.Y.P., Ng, M.-Y., Khong, P.-L., 2020. Lancet Infect. Dis. 20 (4), 384–385.
Letko, M., Marzi, A., Munster, V., 2020. Nature Microbiology 5 (4), 562–569.
Li, F., Li, W., Farzan, M., Harrison, S.C., 2005a. Science 309 (5742), 1864–1868.
Li, W., Zhang, C., Sui, J., Kuhn, J.H., Moore, M.J., Luo, S., Wong, S.-K., Huang, I.-C., Xu, K., Vasilieva, N., Murakami, A., He, Y., Marasco, W.A., Guan, Y., Choe, H., Farzan, M., 2005b. EMBO J. 24 (8), 1634–1643.
Li, Y.-H., Li, J., Liu, X.-E., Wang, L., Li, T., Zhou, Y.-H., Zhuang, H., 2005c. J. Virol Methods 130 (1), 45–50.
Li, Z., Liu, J., Deng, H., Yang, X., Wang, H., Feng, X., Zelinskyy, G., Trilling, M., Sutter, K., Lu, M., Dittmer, U., Wang, B., Yang, D., Zheng, X., Liu, J., 2020a. bioRxiv, 2020.2011.2015.383463.
Li, Z., Yi, Y., Luo, X., Xiong, N., Liu, Y., Li, S., Sun, R., Wang, Y., Hu, B., Chen, W., Zhang, Y., Wang, J., Huang, B., Lin, Y., Yang, J., Cai, W., Wang, X., Cheng, J., Chen, Z., Sun, K., Pan, W., Zhan, Z., Chen, L., Ye, F., 2020b. J. Med. Virol. 92 (9), 1518–1524.
Liao, S.-C., Peng, J., Mauk, M.G., Awasthi, S., Song, J., Friedman, H., Bau, H.H., Liu, C., 2016. Sensor. Actuator. B Chem. 229, 232–238.
Lillehoj, P.B., Huang, M.-C., Truong, N., Ho, C.-M., 2013. Lab Chip 13 (15), 2950–2955.
Liu, et al., 2020. Nature 584 (7821), 450–456.
Liu, W.J., Zhao, M., Liu, K., Xu, K., Wong, G., Tan, W., Gao, G.F., 2017. Antivir. Res. 137, 82–92.
Liu, C., Zhou, Q., Li, Y., Garner, L.V., Watkins, S.P., Carter, L.J., Smoot, J., Gregg, A.C., Daniels, A.D., Jervey, S., Albaiu, D., 2020a. ACS Cent. Sci. 6 (3), 315–331.
Liu, et al., 2020b. EBioMedicine 55, 102763.
Liu, S., Yang, L., Zhang, C., Xiang, Y.-T., Liu, Z., Hu, S., Zhang, B., 2020d. The Lancet Psychiatry 7 (4), e17–e18.
Liu, Y., Liu, Y., Diao, B., Ren, F., Wang, Y., Ding, J., Huang, Q., 2020e. medRxiv, 2020.2003.2026.20044883.
Long, et al., 2020a. Nat. Med. 26 (6), 845–848.
Long, et al., 2020b. Nat. Med. 26 (8), 1200–1204.
Long, et al., 2020c. Nat. Med.
Loynachan, C.N., Thomas, M.R., Gray, E.R., Richards, D.A., Kim, J., Miller, B.S., Brookes, J.C., Agarwal, S., Chudasama, V., McKendry, R.A., Stevens, M.M., 2018. ACS Nano 12 (1), 279–288.
Lu, H., 2020. BioScience Trends 14.
Lu, et al., 2020a. Lancet 395 (10224), 565–574.
Lu, et al., 2020b. Lancet (London, England) 395 (10224), 565–574.
Lv, et al., 2020. BioRxiv, 2020.2003.2015.993097.
Lyu, J., Miao, T., Dong, J., Cao, R., Li, Y., Chen, Q., 2020. Med. Hypotheses 143, 109842-109842.
Mackillop, L., Loerup, L., Bartlett, K., Farmer, A., Gibson, O.J., Hirst, J.E., Kenworthy, Y., Kevat, D.A., Levy, J.C., Tarassenko, L., 2014. Journal of Diabetes Science and Technology 8 (6), 1105–1114.
Madore, D.V., Meade, B.D., Rubin, F., Deal, C., Lynn, F., 2010. Vaccine 28 (29), 4539–4547.
Mahmoudi, T., de la Guardia, M., Baradaran, B., 2020. Trac. Trends Anal. Chem. 125, 115842.
Majors, C.E., Smith, C.A., Natoli, M.E., Kundrod, K.A., Richards-Kortum, R., 2017. Lab Chip 17 (20), 3351–3387.
Majumder, S., Chaudhuri, D., Datta, J., Giri, K., 2020. J. Mol. Graph. Model. 107778.
Malekjahani, A., Sindhwani, S., Syed, A.M., Chan, W.C.W., 2019. Acc. Chem. Res. 52 (9), 2406–2414.
Martel, N., Gomes, S.A., Chemin, I., Trépo, C., Kay, A., 2013. J. Virol Methods 193 (2), 653–659.
Mei, et al., 2020. Nat. Med. 26 (8), 1224–1228.
Menachery, V.D., Dinnon, K.H., Yount, B.L., McAnarney, E.T., Gralinski, L.E., Hale, A., Graham, R.L., Scobey, T., Anthony, S.J., Wang, L., Graham, B., Randell, S.H., Lipkin, W.I., Baric, R.S., 2020. J. Virol. 94 (5) e01774-01719.
Merle, N.S., Noe, R., Halbwachs-Mecarelli, L., Fremeaux-Bacchi, V., Roumenina, L.T., 2015. Front. Immunol. 6, 257-257.
Min, J., Nothing, M., Coble, B., Zheng, H., Park, J., Im, H., Weber, G.F., Castro, C.M., Swirski, F.K., Weissleder, R., Lee, H., 2018. ACS Nano 12 (4), 3378–3384.
Ming, K., Kim, J., Biondi, M.J., Syed, A., Chen, K., Lam, A., Ostrowski, M., Rebbapragada, A., Feld, J.J., Chan, W.C.W., 2015. ACS Nano 9 (3), 3060–3074.
Miripour, Z.S., Sarrami-Forooshani, R., Sanati, H., Makarem, J., Taheri, M.S., Shojaeian, F., Eskafi, A.H., Abbasvandi, F., Namdar, N., Ghafari, H., Aghaee, P., Zandi, A., Faramarzpour, M., Hoseinyazdi, M., Tayebi, M., Abdolahad, M., 2020. Biosens. Bioelectron. 165, 112435.
Mizumoto, K., Kagaya, K., Zarebski, A., Chowell, G., 2020. Estimating the asymptomatic proportion of coronavirus disease 2019 (COVID-19) cases on board the Diamond Princess cruise ship, Yokohama, Japan. Euro Surveill. 25 (10), 2000180, 2020.
Mollaei, H.R., Afshar, A.A., Kalantar-Neyestanaki, D., Fazlalipour, M., Aflatoonian, B., 2020. Iran. J. Microbiol. 12 (3), 185–193.
Montesinos, I., Gruson, D., Kabamba, B., Dahma, H., Van den Wijngaert, S., Reza, S., Carbone, V., Vandenberg, O., Gulbis, B., Wolff, F., Rodriguez-Villalobos, H., 2020. J. Clin. Virol. 128, 104413.
Moore, J.B., June, C.H., 2020. Science 368 (6490), 473–474.
Morales-Narváez, E., Dincer, C., 2020. Biosens. Bioelectron. 163, 112274.
Mori, Y., Nagamine, K., Tomita, N., Notomi, T., 2001. Biochem. Biophys. Res. Commun. 289 (1), 150–154.
Mousavizadeh, L., Ghasemi, S., 2020. J. Microbiol. Immunol. Infect. 1620, 30082–30087. S1684-1182.
Mudanyali, O., Dimitrov, S., Sikora, U., Padmanabhan, S., Navruz, I., Ozcan, A., 2012a. Lab Chip 12 (15), 2678–2686.
Mudanyali, O., Dimitrov, S., Sikora, U., Padmanabhan, S., Navruz, I., Ozcan, A., 2012b. Lab Chip 12 (15), 2678–2686.
Nachtigall, F.M., Pereira, A., Trofymchuk, O.S., Santos, L.S., 2020. Nat. Biotechnol. 38 (10), 1168–1173.
Nakajima, S., Hoshina, H., Yamashita, M., Otani, C., Miyoshi, N., 2007. Appl. Phys. Lett. 90 (4), 041102.
Nayak, S., Blumenfeld, N.R., Laksanasopin, T., Sia, S.K., 2017. Anal. Chem. 89 (1), 102–123.
Neuman, B.W., Kiss, G., Kunding, A.H., Bhella, D., Baksh, M.F., Connelly, S., Droese, B., Klaus, J.P., Makino, S., Sawicki, S.G., Siddell, S.G., Stamou, D.G., Wilson, I.A., Kuhn, P., Buchmeier, M.J., 2011. J. Struct. Biol. 174 (1), 11–22.
Ng, et al., 2018. Sci. Transl. Med. 10 (438) eaar6076.
Nilsson, H.-O., Aleljung, P., Nilsson, I., Tyszkiewicz, T., Wadström, T., 1996. J. Microbiol. Methods 27 (1), 73–79.
Notomi, T., Okayama, H., Masubuchi, H., Yonekawa, T., Watanabe, K., Amino, N., Hase, T., 2000. Nucleic Acids Res. 28 (12) e63-e63.
Nuccetelli, M., Pieri, M., Grelli, S., Ciotti, M., Miano, R., Andreoni, M., Bernardini, S., 2020. Cell Death Discovery 6 (1), 38.
Oja, A.E., Saris, A., Ghandour, C.A., Kragten, N.A.M., Hogema, B.M., Nossent, E.J., Heunks, L.M.A., Cuvalay, S., Slot, E., Swaneveld, F.H., Vrielink, H., Rispens, T., van der Schoot, E., van Lier, R.A.W., Brinke, A.T., Hombrink, P., 2020. BioRxiv, 2020.2006.2018.159202.
Okba, N.M.A., Müller, M., Li, W., Wang, C., GeurtsvanKessel, C., Corman, V., Lamers, M., Sikkema, R., de Bruin, E., Chandler, F., Yazdanpanah, Y., Le Hingrat, Q., Descamps, D., Houhou-Fidouh, N., Reusken, C.B.E.M., Bosch, B.-J., Drosten, C., Koopmans, M.P.G., Haagmans, B., 2020. Emerging Infectious Disease journal 26 (7), 1478.
Oliver, N., Lepri, B., Sterly, H., Lambiotte, R., Deletaille, S., De Nadai, M., Letouzé, E., Salah, A.A., Benjamins, R., Cattuto, C., Colizza, V., de Cordes, N., Fraiberger, S.P., Koebe, T., Lehmann, S., Murillo, J., Pentland, A., Pham, P.N., Pivetta, F., Saramäki, J., Scarpino, S.V., Tizzoni, M., Verhulst, S., Vinck, P., 2020. Science Advances 6 (23) eabc0764.
Orooji, Y., Sohrabi, H., Hemmat, N., Oroojalian, F., Baradaran, B., Mokhtarzadeh, A., Mohaghegh, M., Karimi-Maleh, H., 2020. Nano-Micro Lett. 13 (1), 18.
Ou, X., Liu, Y., Lei, X., Li, P., Mi, D., Ren, L., Guo, L., Guo, R., Chen, T., Hu, J., Xiang, Z., Mu, Z., Chen, X., Chen, J., Hu, K., Jin, Q., Wang, J., Qian, Z., 2020. Nat. Commun. 11 (1), 1620.
Padoan, A., Cosma, C., Sciacovelli, L., Faggian, D., Plebani, M., 2020. Clin. Chem. Lab. Med. 58 (7), 1081.
Pal, M., Berhanu, G., Desalegn, C., Kandi, V., 2020. Cureus 12 (3) e7423-e7423.
Pallesen, J., Wang, N., Corbett, K.S., Wrapp, D., Kirchdoerfer, R.N., Turner, H.L., Cottrell, C.A., Becker, M.M., Wang, L., Shi, W., Kong, W.-P., Andres, E.L., Kettenbach, A.N., Denison, M.R., Chappell, J.D., Graham, B.S., Ward, A.B., McLellan, J.S., 2017. Proc. Natl. Acad. Sci. Unit. States Am. 114 (35), E7348–E7357.
Pan, D., Sze, S., Minhas, J.S., Bangash, M.N., Pareek, N., Divall, P., Williams, C.M.L., Oggioni, M.R., Squire, I.B., Nellums, L.B., Hanif, W., Khunti, K., Pareek, M., 2020a. EClinicalMedicine 23, 100404.
Pan, F., Ye, T., Sun, P., Gui, S., Liang, B., Li, L., Zheng, D., Wang, J., Hesketh, R.L., Yang, L., Zheng, C., 2020b. Radiology 295 (3), 715–721.
Pan, Y., Zhang, D., Yang, P., Poon, L.L.M., Wang, Q., 2020c. Lancet Infect. Dis. 20 (4), 411–412.
Park, H.-R., Chen, X., Nguyen, N.-C., Peraire, J., Oh, S.-H., 2015. ACS Photonics 2 (3), 417–424.
Park, M., Won, J., Choi, B.Y., Lee, C.J., 2020. Exp. Mol. Med. 52 (6), 963–977.
Patchsung, et al., 2020. Nature Biomedical Engineering.
Peeling, R.W., Mabey, D., 2010. Clin. Microbiol. Infect. 16 (8), 1062–1069.
Pereira, et al., 2020. Genet. Med. 22 (9), 1467–1469.
Perkel, J.M., 2017. Nature 545 (7652), 119–121.
Petryayeva, E., Algar, W.R., 2015. Analyst 140 (12), 4037–4045.
Pinto, et al., 2020. Nature 583 (7815), 290–295.
Pisani, E., Aaby, P., Breugelmans, J.G., Carr, D., Groves, T., Helinski, M., Kamuya, D., Kern, S., Littler, K., Marsh, V., Mboup, S., Merson, L., Sankoh, O., Serafini, M., Schneider, M., Schoenenberger, V., Guerin, P.J., 2016. BMJ 355, i5295.
Pondaven-Letourmy, S., Alvin, F., Boumghit, Y., Simon, F., 2020. Eur Ann Otorhinolaryngol Head Neck Dis 137 (4), 325–327.
Prilutskiy, A., Kritselis, M., Shevtsov, A., Yambayev, I., Vadlamudi, C., Zhao, Q., Kataria, Y., Sarosiek, S.R., Lerner, A., Sloan, J.M., Quillen, K., Burks, E.J., 2020. Am. J. Clin. Pathol. 154 (4), 466–474.
Priye, A., Bird, S.W., Light, Y.K., Ball, C.S., Negrete, O.A., Meagher, R.J., 2017. Sci. Rep. 7 (1), 44778.
Quinlan, B.D., Mou, H., Zhang, L., Guo, Y., He, W., Ojha, A., Parcells, M.S., Luo, G., Li, W., Zhong, G., Choe, H., Farzan, M., 2020. BioRxiv, 2020.2004.2010.036418.
Rajan, N., Joshi, G.P., 2020. COVID-19: Anesth. Analg. 131 (1), 31–36.
Rastogi, Y.R., Sharma, A., Nagraik, R., Aygün, A., Şen, F., 2020. Int. J. Environ. Sci. Technol.
Ravi, N., Cortade, D.L., Ng, E., Wang, S.X., 2020. Biosens. Bioelectron. 165, 112454.
Reguera, J., Mudgal, G., Santiago, C., Casasnovas, J.M., 2014. Virus Res. 194, 3–15.

Richard, M., Kok, A., de Meulder, D., Bestebroer, T.M., Lamers, M.M., Okba, N.M.A., Fentener van Vlissingen, M., Rockx, B., Haagmans, B.L., Koopmans, M.P.G., Fouchier, R.A.M., Herfst, S., 2020. Nat. Commun. 11 (1), 3496.
Rissin, D.M., Kan, C.W., Campbell, T.G., Howes, S.C., Fournier, D.R., Song, L., Piech, T., Patel, P.P., Chang, L., Rivnak, A.J., Ferrell, E.P., Randall, J.D., Provuncher, G.K., Walt, D.R., Duffy, D.C., 2010. Nat. Biotechnol. 28 (6), 595–599.
Robbiani, et al., 2020. Nature 584 (7821), 437–442.
Robson, B., 2020a. Comput. Biol. Med. 119, 103670-103670.
Robson, B., 2020b. Comput. Biol. Med. 103749.
Rocca, M.F., Zintgraff, J.C., Dattero, M.E., Santos, L.S., Ledesma, M., Vay, C., Prieto, M., Benedetti, E., Avaro, M., Russo, M., Nachtigall, F.M., Baumeister, E., 2020. J. Virol Methods 286, 113991-113991.
Rong, Z., Wang, Q., Sun, N., Jia, X., Wang, K., Xiao, R., Wang, S., 2019. Anal. Chim. Acta 1055, 140–147.
Rowe, T., Abernathy, R.A., Hu-Primmer, J., Thompson, W.W., Lu, X., Lim, W., Fukuda, K., Cox, N.J., Katz, J.M., 1999. J. Clin. Microbiol. 37 (4), 937–943.
Rubin, et al., 2020. Radiology 296 (1), 172–180.
Rumbold, B., Wenham, C., Wilson, J., 2017. BMC Med. Ethics 18 (1), 33.
Ryu, S., Ali, S.T., Lim, J.-S., Chun, B.C., 2020. Int. J. Environ. Res. Publ. Health 17 (9), 3113.
Saif, L.J., 2004. Rev Sci Tech 23 (2), 643–660.
Salam, N., Rane, S., Das, R., Faulkner, M., Gund, R., Kandpal, U., Lewis, V., Mattoo, H., Prabhu, S., Ranganathan, V., Durdik, J., George, A., Rath, S., Bal, V., 2013. Indian J. Med. Res. 138 (5), 595–608.
Sanyaolu, A., Okorie, C., Marinkovic, A., Ayodele, O., Abbasi, A.F., Prakash, S., Ahmed, M., Kayode, D., Jaferi, U., Haider, N., 2020. SN Comprehensive Clinical Medicine 2 (9), 1393–1400.
Schoeman, D., Fielding, B.C., 2019. Virol. J. 16 (1), 69.
Schröder, I., 2020. ACS Chemical Health & Safety.
Sekine, et al., 2020. BioRxiv, 2020.2006.2029.174888.
Seo, et al., 2009. Nat. Photon. 3 (3), 152–156.
Seo, et al., 2020. ACS Nano 14 (4), 5135–5142.
Sepe, O., Valiani, R., Fasanaro, A.M., Mandarini, A., Verdoliva, A., 1981. Acta Neurol Quad (Napoli) 42, 157–162.
Shang, J., Wan, Y., Luo, C., Ye, G., Geng, Q., Auerbach, A., Li, F., 2020a. Proc. Natl. Acad. Sci. U.S.A. 117 (21), 11727–11734.
Shang, J., Ye, G., Shi, K., Wan, Y., Luo, C., Aihara, H., Geng, Q., Auerbach, A., Li, F., 2020b. Nature 581 (7807), 221–224.
Sheahan, T., Rockx, B., Donaldson, E., Sims, A., Pickles, R., Corti, D., Baric, R., 2008. J. Virol. 82 (5), 2274–2285.
Shen, et al., 2020. Am J Transl Res 12 (4), 1348–1354.
Shereen, M.A., Khan, S., Kazmi, A., Bashir, N., Siddique, R., 2020. J. Adv. Res. 24, 91–98.
Shi, et al., 2020. Science (New York, N.Y.) 368 (6494), 1016–1020.
Shin, H.S., Kim, Y., Kim, G., Lee, J.Y., Jeong, I., Joh, J.S., Kim, H., Chang, E., Sim, S.Y., Park, J.S., Lim, D.G., 2019. Clin. Infect. Dis. 68 (6), 984–992.
Shirato, K., Nishimura, H., Saijo, M., Okamoto, M., Noda, M., Tashiro, M., Taguchi, F., 2007. J. Virol Methods 139 (1), 78–84.
Shrivastava, S., Lee, W.I., Lee, N.E., 2018. CBiosensors & Bioelectronics 109, 90–97.
Smyrlaki, I., Ekman, M., Lentini, A., Rufino de Sousa, N., Papanicolaou, N., Vondracek, M., Aarum, J., Safari, H., Muradrasoli, S., Rothfuchs, A.G., Albert, J., Högberg, B., Reinius, B., 2020. Nat. Commun. 11 (1), 4812.
Snodgrass, R., Gardner, A., Semeere, A., Kopparthy, V.L., Duru, J., Maurer, T., Martin, J., Cesarman, E., Erickson, D., 2018. Nature Biomedical Engineering 2 (9), 657–665.
Sobolev, et al., 2016. Nat. Immunol. 17 (2), 204–213.
Song, W., Gui, M., Wang, X., Xiang, Y., 2018. PLoS Pathog. 14 (8), e1007236.
Song, Y., Song, J., Wei, X., Huang, M., Sun, M., Zhu, L., Lin, B., Shen, H., Zhu, Z., Yang, C., 2020. Anal. Chem. 92 (14), 9895–9900.
Sonnberg, S., Webby, R.J., Webster, R.G., 2013. Virus Res. 178 (1), 63–77.
Søreide, K., Hallet, J., Matthews, J.B., Schnitzbauer, A.A., Line, P.D., Lai, P.B.S., Otero, J., Callegaro, D., Warner, S.G., Baxter, N.N., Teh, C.S.C., Ng-Kamstra, J., Meara, J.G., Hagander, L., Lorenzon, L., 2020. Br. J. Surg. 107 (10), 1250–1261.
Spengler, M., Adler, M., Niemeyer, C.M., 2015. Analyst 140 (18), 6175–6194.
Spinello, A., Saltalamacchia, A., Magistrato, A., 2020. J. Phys. Chem. Lett. 11 (12), 4785–4790.
Stedtfeld, R.D., Tourlousse, D.M., Seyrig, G., Stedtfeld, T.M., Kronlein, M., Price, S., Ahmad, F., Gulari, E., Tiedje, J.M., Hashsham, S.A., 2012. Lab Chip 12 (8), 1454–1462.
Sun, H., Dickens, B.L., Chen, M., Cook, A.R., Clapham, H.E., 2020. MedRxiv, 2020.2002.2017.20024075.
Swystun, L.L., Liaw, P.C., 2016. Blood 128 (6), 753–762.
Tai, W., He, L., Zhang, X., Pu, J., Voronin, D., Jiang, S., Zhou, Y., Du, L., 2020. Cell. Mol. Immunol. 17 (6), 613–620.
Tan, J., Liu, S., Zhuang, L., Chen, L., Dong, M., Zhang, J., Xin, Y., 2020a. Future Virol. https://doi.org/10.2217/fvl-2020-0087.
Tan, X., Krel, M., Dolgov, E., Park, S., Li, X., Wu, W., Sun, Y.-L., Zhang, J., Khaing Oo, M. K., Perlin, D.S., Fan, X., 2020b. Biosens. Bioelectron. 169, 112572-112572.
Tan, X., Krel, M., Dolgov, E., Park, S., Li, X., Wu, W., Sun, Y.-L., Zhang, J., Khaing Oo, M. K., Perlin, D.S., Fan, X., 2020c. Biosens. Bioelectron. 169, 112572.
Tang, X., Wu, C., Li, X., Song, Y., Yao, X., Wu, X., Duan, Y., Zhang, H., Wang, Y., Qian, Z., Cui, J., Lu, J., 2020. National Science Review 7 (6), 1012–1023.
Tenggara, A.P., Park, S.J., Yudistira, H.T., Ahn, Y.H., Byun, D., 2017. J. Micromech. Microeng. 27 (3), 035009.
Thaxton, C.S., Elghanian, R., Thomas, A.D., Stoeva, S.I., Lee, J.-S., Smith, N.D., Schaeffer, A.J., Klocker, H., Horninger, W., Bartsch, G., Mirkin, C.A., 2009. Proc. Natl. Acad. Sci. Unit. States Am. 106 (44), 18437–18442.
Thwe, P.M., Ren, P., 2020. Diagn. Microbiol. Infect. Dis. 115228.
Tian, X., Li, C., Huang, A., Xia, S., Lu, S., Shi, Z., Lu, L., Jiang, S., Yang, Z., Wu, Y., Ying, T., 2020. Emerg. Microb. Infect. 9 (1), 382–385.
Tilocca, B., Soggiu, A., Sanguinetti, M., Musella, V., Britti, D., Bonizzi, L., Urbani, A., Roncada, P., 2020. Microb. Infect. 22 (4), 188–194.
To, et al., 2020. Lancet Infect. Dis. 20 (5), 565–574.
Tortorici, M.A., Veesler, D., 2019. Adv. Virus Res. 105, 93–116.
Tresp, V., Overhage, J.M., Bundschus, M., Rabizadeh, S., Fasching, P.A., Yu, S., 2016. Proc. IEEE 104 (11), 2180–2206.
Turbé, V., Gray, E.R., Lawson, V.E., Nastouli, E., Brookes, J.C., Weiss, R.A., Pillay, D., Emery, V.C., Verrips, C.T., Yatsuda, H., Athey, D., McKendry, R.A., 2017. Sci. Rep. 7 (1), 11971.
Udugama, B., Kadhiresan, P., Kozlowski, H.N., Malekjahani, A., Osborne, M., Li, V.Y.C., Chen, H., Mubareka, S., Gubbay, J.B., Chan, W.C.W., 2020. ACS Nano 14 (4), 3822–3835.
Vabret, et al., 2020. Immunity 52 (6), 910–941.
van Kasteren, P.B., van der Veer, B., van den Brink, S., Wijsman, L., de Jonge, J., van den Brandt, A., Molenkamp, R., Reusken, C.B.E.M., Meijer, A., 2020. J. Clin. Virol. 128, 104412.
Vandenberg, O., Martiny, D., Rochas, O., van Belkum, A., Kozlakidis, Z., 2020. Nat. Rev. Microbiol.
Varadé, J., Magadán, S., González-Fernández, Á., 2020. Cell. Mol. Immunol.
Walls, A.C., Park, Y.-J., Tortorici, M.A., Wall, A., McGuire, A.T., Veesler, D., 2020. Cell 181 (2), 281–292 e286.
Wan, Y., Shang, J., Graham, R., Baric, R.S., Li, F., 2020. J. Virol. 94 (7) e00127-00120.
Wang, S., Zhao, X., Khimji, I., Akbas, R., Qiu, W., Edwards, D., Cramer, D.W., Ye, B., Demirci, U., 2011a. Lab Chip 11 (20), 3411–3418.
Wang, S., Zhao, X., Khimji, I., Akbas, R., Qiu, W., Edwards, D., Cramer, D.W., Ye, B., Demirci, U., 2011b. Lab Chip 11 (20), 3411–3418.
Wang, C., Li, W., Drabek, D., Okba, N.M.A., van Haperen, R., Osterhaus, A.D.M.E., van Kuppeveld, F.J.M., Haagmans, B.L., Grosveld, F., Bosch, B.-J., 2020a. Nat. Commun. 11 (1), 2251.
Wang, X., Xiong, E., Tian, T., Cheng, M., Lin, W., Wang, H., Zhang, G., Sun, J., Zhou, X., 2020b. ACS Nano 14 (2), 2497–2508.
Wat, D., Gelder, C., Hibbitts, S., Cafferty, F., Bowler, I., Pierrepoint, M., Evans, R., Doull, I., 2008. J. Cyst. Fibros. 7 (4), 320–328.
Wei, Q., Qi, H., Luo, W., Tseng, D., Ki, S.J., Wan, Z., Göröcs, Z., Bentolila, L.A., Wu, T.-T., Sun, R., Ozcan, A., 2013. ACS Nano 7 (10), 9147–9155.
Weiskopf, D., Schmitz, K.S., Raadsen, M.P., Grifoni, A., Okba, N.M.A., Endeman, H., van den Akker, J.P.C., Molenkamp, R., Koopmans, M.P.G., van Gorp, E.C.M., Haagmans, B.L., de Swart, R.L., Sette, A., de Vries, R.D., 2020. Sci. Immunol. 5 (48).
Whiting, P., Singatullina, N., Rosser, J.H., 2015. BJA Education 15 (6), 299–304.
Wilk, A.J., Rustagi, A., Zhao, N.Q., Roque, J., Martínez-Colón, G.J., McKechnie, J.L., Ivison, G.T., Ranganath, T., Vergara, R., Hollis, T., Simpson, L.J., Grant, P., Subramanian, A., Rogers, A.J., Blish, C.A., 2020. Nat. Med. 26 (7), 1070–1076.
Winichakoon, P., Chaiwarith, R., Liwsrisakun, C., Salee, P., Goonna, A., Limsukon, A., Kaewpoowat, Q., 2020. J. Clin. Microbiol. 58 (5) e00297-00220.
Wölfel, et al., 2020. Nature 581 (7809), 465–469.
Wong, M.L., Medrano, J.F., 2005. Biotechniques 39 (1), 75–85.
Wood, C.S., Thomas, M.R., Budd, J., Mashamba-Thompson, T.P., Herbst, K., Pillay, D., Peeling, R.W., Johnson, A.M., McKendry, R.A., Stevens, M.M., 2019. Nature 566 (7745), 467–474.
Wrapp, D., Wang, N., Corbett, K.S., Goldsmith, J.A., Hsieh, C.-L., Abiona, O., Graham, B. S., McLellan, J.S., 2020. Science 367 (6483), 1260–1263.
Wu, A., Peng, Y., Huang, B., Ding, X., Wang, X., Niu, P., Meng, J., Zhu, Z., Zhang, Z., Wang, J., Sheng, J., Quan, L., Xia, Z., Tan, W., Cheng, G., Jiang, T., 2020a. Cell Host Microbe 27 (3), 325–328.
Wu, F., Zhao, S., Yu, B., Chen, Y.-M., Wang, W., Song, Z.-G., Hu, Y., Tao, Z.-W., Tian, J.-H., Pei, Y.-Y., Yuan, M.-L., Zhang, Y.-L., Dai, F.-H., Liu, Y., Wang, Q.-M., Zheng, J.-J., Xu, L., Holmes, E.C., Zhang, Y.-Z., 2020b. Nature 579 (7798), 265–269.
Xia, S., Liu, M., Wang, C., Xu, W., Lan, Q., Feng, S., Qi, F., Bao, L., Du, L., Liu, S., Qin, C., Sun, F., Shi, Z., Zhu, Y., Jiang, S., Lu, L., 2020. Cell Res. 30 (4), 343–355.
Xiang, F., Wang, X., He, X., Peng, Z., Yang, B., Zhang, J., Zhou, Q., Ye, H., Ma, Y., Li, H., Wei, X., Cai, P., Ma, W.-L., 2020a. Clin. Infect. Dis. 71 (8), 1930–1934.
Xiang, J., Yan, M., Li, H., Liu, T., Lin, C., Huang, S., Shen, C., 2020b. MedRxiv, 2020.2002.2027.20028787.
Xie, X., Zhong, Z., Zhao, W., Zheng, C., Wang, F., Liu, J., 2020. Radiology 296 (2), E41–E45.
Xu, M., Wang, D., Wang, H., Zhang, X., Liang, T., Dai, J., Li, M., Zhang, J., Zhang, K., Xu, D., Yu, X., 2020. Clin. Transl. Med. 10 (4), e158.
Yan, R., Zhang, Y., Li, Y., Xia, L., Guo, Y., Zhou, Q., 2020. Science 367 (6485), 1444–1448.
Yang, W., Yan, F., 2020. Radiology 295 (2). E3-E3.
Yang, et al., 2020a. Nature 586 (7830), 572–577.
Yang, L., Liu, S., Liu, J., Zhang, Z., Wan, X., Huang, B., Chen, Y., Zhang, Y., 2020b. Signal Transduct. Target. Ther. 5 (1), 128.
Yang, W., Dang, X., Wang, Q., Xu, M., Zhao, Q., Zhou, Y., Zhao, H., Wang, L., Xu, Y., Wang, J., Han, S., Wang, M., Pei, F., Wang, Y., 2020c. MedRxiv, 2020.2003.2002.20030130.
Yang, Y., Peng, F., Wang, R., Guan, K., Jiang, T., Xu, G., Sun, J., Chang, C., 2020d. J. Autoimmun. 109, 102434-102434.
Yarmarkovich, M., Warrington, J.M., Farrel, A., Maris, J.M., 2020. Cell Rep. Med. 1 (3), 100036.
Ye, X., Li, L., Li, J., Wu, X., Fang, X., Kong, J., 2019. ACS Sens. 4 (11), 3066–3071.
Ye, Q., Wang, B., Mao, J., 2020. J. Infect. 80 (6), 607–613.

Yeo, S.-J., Choi, K., Cuc, B.T., Hong, N.N., Bao, D.T., Ngoc, N.M., Le, M.Q., Hang, N.L.K., Thach, N.C., Mallik, S.K., Kim, H.S., Chong, C.-K., Choi, H.S., Sung, H.W., Yu, K., Park, H., 2016. Theranostics 6 (2), 231–242.
Yi, C., Sun, X., Ye, J., Ding, L., Liu, M., Yang, Z., Lu, X., Zhang, Y., Ma, L., Gu, W., Qu, A., Xu, J., Shi, Z., Ling, Z., Sun, B., 2020. Cell. Mol. Immunol. 17 (6), 621–630.
Yong, C.Y., Ong, H.K., Yeap, S.K., Ho, K.L., Tan, W.S., 2019. Front. Microbiol. 10, 1781.
Yu, L., Wu, S., Hao, X., Li, X., Liu, X., Ye, S., Han, H., Dong, X., Li, X., Li, J., Liu, J., Liu, N., Zhang, W., Pelechano, V., Chen, W.-H., Yin, X., 2020a. MedRxiv, 2020.2002.2020.20025874.
Yu, M., Xu, D., Lan, L., Tu, M., Liao, R., Cai, S., Cao, Y., Xu, L., Liao, M., Zhang, X., Xiao, S.-Y., Li, Y., Xu, H., 2020b. Radiology: Cardiothoracic Imaging 2 (2), e200126.
Yuan, Z., Chen, Y.-C., Li, H.-W., Chang, H.-T., 2014. Chem. Commun. 50 (69), 9800–9815.
Zhang, B.-Z., Chu, H., Han, S., Shuai, H., Deng, J., Hu, Y.-f., Gong, H.-r., Lee, A.C.-Y., Zou, Z., Yau, T., Wu, W., Hung, I.F.-N., Chan, J.F.-W., Yuen, K.-Y., Huang, J.-D., 2020a. Cell Res.
Zhang, G., Pomplun, S., Loftis, A.R., Tan, X., Loas, A., Pentelute, B.L., 2020b. BioRxiv, 2020.2003.2019.999318.
Zhang, R., Liu, H., Li, F., Zhang, B., Liu, Q., Li, X., Luo, L., 2020c. MedRxiv, 2020.2001.2030.20019836.
Zhang, T., Wu, Q., Zhang, Z., 2020d. Curr. Biol. 30 (7), 1346–1351 e1342.
Zhang, W., Du, R.-H., Li, B., Zheng, X.-S., Yang, X.-L., Hu, B., Wang, Y.-Y., Xiao, G.-F., Yan, B., Shi, Z.-L., Zhou, P., 2020e. Emerg. Microb. Infect. 9 (1), 386–389.
Zhang, Y., Odiwuor, N., Xiong, J., Sun, L., Nyaruaba, R.O., Wei, H., Tanner, N.A., 2020f. medRxiv, 2020.2002.2026.20028373.
Zhao, Y., Chen, F., Li, Q., Wang, L., Fan, C., 2015. Chem. Rev. 115 (22), 12491–12545.
Zhao, V.X.T., Wong, T.I., Zheng, X.T., Tan, Y.N., Zhou, X., 2020. Mater. Sci. Energy Technol. 3, 237–249.
Zheng, J., 2020. Int. J. Biol. Sci. 16 (10), 1678–1685.
Zheng, M., Song, L., 2020. Cell. Mol. Immunol. 17 (5), 536–538.
Zheng, M., Gao, Y., Wang, G., Song, G., Liu, S., Sun, D., Xu, Y., Tian, Z., 2020a. Cell. Mol. Immunol. 17 (5), 533–535.
Zheng, Z., Yao, Z., Wu, K., Zheng, J., 2020b. J. Med. Virol. 92 (11), 2420–2428.
Zhou, F., Yu, T., Du, R., Fan, G., Liu, Y., Liu, Z., Xiang, J., Wang, Y., Song, B., Gu, X., Guan, L., Wei, Y., Li, H., Wu, X., Xu, J., Tu, S., Zhang, Y., Chen, H., Cao, B., 2020a. Lancet 395 (10229), 1054–1062.
Zhou, et al., 2020b. Nature 579 (7798), 270–273.
Zhou, S., Zhu, T., Wang, Y., Xia, L., 2020c. Eur. Radiol. 1–9.
Zhou, T., Huang, R., Huang, M., Shen, J., Shan, Y., Xing, D., 2020d. Advanced Science 7 (13), 1903661.
Zhu, N., Zhang, D., Wang, W., Li, X., Yang, B., Song, J., Zhao, X., Huang, B., Shi, W., Lu, R., Niu, P., Zhan, F., Ma, X., Wang, D., Xu, W., Wu, G., Gao, G.F., Tan, W., 2020a. N. Engl. J. Med. 382 (8), 727–733.
Zhu, X., Wang, X., Han, L., Chen, T., Wang, L., Li, H., Li, S., He, L., Fu, X., Chen, S., Xing, M., Chen, H., Wang, Y., 2020b. Biosens. Bioelectron. 166, 112437.