doi.org/10.36721/PJPS.2021.34.5.REG.1813-1820.1

REVIEW

Covid-19: Diagnosis, summary of essays and evolving approaches

Muhammad Usman Munir[1]*, Sajal Salman[2], Arsalan Ahmed[3], Waqar Iqbal[4], Syed Nasir Abbas Bukhari[1], Naveed Ahmad[5], Muhammad Ikram Ullah[6], Muhammad Masood Ahmad[5], Tilal Elsaman[1,7] and Muhammad Ali Tahir[8]

[1]Department of Pharmaceutical Chemistry, College of Pharmacy, Jouf University, Sakaka, Aljouf, Saudi Arabia
[2]Faculty of Pharmacy, University of Central Punjab, Lahore, Pakistan
[3]Interdisciplinary Research Centre in Biomedical Materials, COMSATS Institute of Information Technology, Lahore, Pakistan
[4]School of Textile Science and Engineering, Tiangong University, Tianjin, China
[5]Department of Pharmaceutics, College of Pharmacy, Jouf University, Sakaka, Aljouf, Saudi Arabia
[6]Department of Clinical Laboratory Sciences, Jouf University, Sakaka, Aljouf, Saudi Arabia
[7]Department of Pharmaceutical Chemistry, College of Pharmacy, Omdurman Islamic University, Khartoum, Sudan
[8]Shanghai Key Laboratory of Atmospheric Particle Pollution and Prevention, Department of Environmental Science & Engineering, Fudan University, Shanghai, Peoples' Republic of China

Abstract: COVID-19 spread worldwide after its outbreak in December 2019. This review paper aims to educate the readers regarding SARS-CoV-2 diagnostic and detection tools and the issues experienced by researchers. We identify on-the-horizon point-of-care diagnostic tests and inspire scholars to develop their innovations past conception. It will also effectively avoid potential pandemics to establish plug-and-play diagnostic information to handle the SARS infection. The authors agree that arbitrary-access, interconnected systems with flexible functionality accessible at the point-of-care, would enable fast and precise diagnosis and tracking.

Keywords: COVID-19, SARS-CoV-2, diagnostic tests, smartphone surveillance, point-of-care.

INTRODUCTION

The patients exposed to COVID-19 were confessed with high temperature, coughing, lack of breath, as well as various other signs and symptoms (Zhou *et al.*, 2020; Hamid, Masood, Tariq, *et al.*, 2020). People were checked to use a computed tomography (CT), which disclosed different opacities compared to pictures of healthy and balanced lung (Ai *et al.*, 2020). It leads to the preliminary medical diagnosis of pneumonia. Supplementary nucleic acid evaluation utilizing complex real-time polymerase domino effect (PCR) of understood microorganism panels caused unfavorable outcomes. It is recommended that the pneumonia cause is of the unidentified source (Chauhan *et al.*, 2020). The samples were taken from the patients' bronchoalveolar lavage (BAL) liquid by January 10, 2020. They were sequenced to expose a virus having comparable hereditary series to the betacoronavirus B family tree. The researchers found that this brand-new microorganism has ~ 80%, ~ 50%, as well as ~ 96% resemblance to the genome of the Middle East respiratory syndrome (MERS-CoV), severe acute respiratory syndrome (SARS-CoV), and also bat coronavirus RaTG13, precisely (R. Lu *et al.*, 2020; Ji *et al.*, 2020). The unique coronavirus was called SARS-CoV-2, the virus triggering COVID-19. Since September 30, 2020, it has actually infected a minimum of 215 nations, contaminated 33,865,157 individuals, and has actually caused 1,012,940 fatalities worldwide. It is thought that the complete variety of testified COVID-19 infections is ignored as there are several light or asymptomatic instances (Kobayashi *et al.*, 2020). Ruby Princess cruise liner research reported a quote of 17.9% of asymptomatic situations were reported. The people with no symptoms are as contagious as symptomatic people as well as are consequently efficient in adding to more spreading out the illness (Mizumoto *et al.*, 2020).

SARS-CoV-2 can be directed from humans to humans (Joshi *et al.*, 2020). It is approximated that a SARS-CoV-2-infected individual will certainly contaminate around three brand-new individuals. The viral structure of SARS-COV-2 is shown in fig. 1. The signs and indications can differ, with some people continuing to be asymptomatic while others are existing with high temperature, coughing, exhaustion, as well as a host of various other signs and symptoms (Oran and Topol, 2020). The signs might resemble people with flu or cold. At this phase, one of the probable settings of transmission is believed to be via straight call and also droplet-spread (Li *et al.*, 2020). Current research is taking a look at aerosol, and also surface area security of SARS-CoV-2 revealed that the infection might be located in aerosols (<5μm) at the very least as much as 3 h and also might be extra steady on

*Corresponding author: e-mail: mumunir@ju.edu.sa

plastic as well as stainless-steel than on copper as well as cardboard (Van Doremalen *et al.*, 2020).

The vaccine for COVID-19 is under development process and at this stage, there is no USA-FDA approved vaccine in the market (Liu *et al.*, 2020; Hamid, Masood, Khalid, *et al.*, 2020). The diagnostic tools have a crucial duty in the control of COVID-19, allowing the fast application of control procedures that restrict the spread with situation recognition, seclusion, as well as call mapping (i.e., recognizing individuals that might have can be found in contact with a contaminated client). The COVID-19 workflow for diagnosis is present in fig. 2. Herein, we intend to sum up the present well-known organic residential or commercial properties of SARS-CoV-2, analysis devices, and scientific outcomes for identifying SARS-CoV-2, arising diagnostics, and security innovation to suppress the spread. It is currently a hot topic in the research that incorporates the present updates that might work for assisting techniques to manage a prospective COVID-19 pandemic.

Diagnostic techniques for COVID-19

The US's public health officials reported that an excessive increase in cases of COVID-19 highlighted the significance of in-vitro analysis of ailments like SARS-CoV-2 (Di Gennaro *et al.*, 2020). The COVID-19 infectious patients show vague symptoms that cannot be utilized for a precise finding. The researchers presented that among 1099 cases in China, around 44% of patients had a fever when they got hospitalized, whereas about 89% of patients got a fever while already being hospitalized (Fu *et al.*, 2020). Moreover, it was observed that around 19% of patients had dyspnea, 34% of cases had sputum production, 38% of patients displayed fatigue, and about 68% of cases presented a cough. Some symptoms might be related to other respiratory diseases (Verma *et al.,* 2020; Wang *et al.*, 2020).

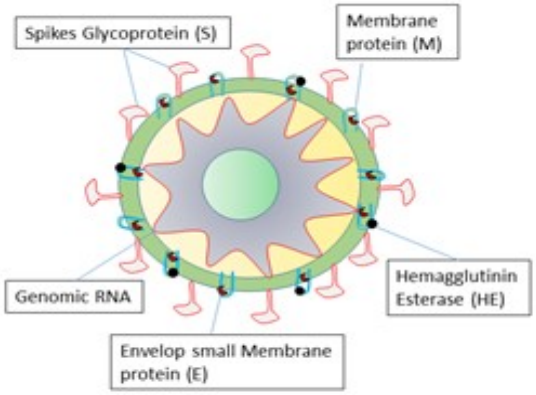

Fig. 1: Illustration of SARS-CoV-2 structure.

Computed tomography (CT) and nucleic acid analysis are employed for COVID-19 detection. CT and molecular methods can detect and recognize specific pathogens; thus, these are considered appropriate for precise diagnosis. The development of molecular methods depends on the genetic and protein framework of the pathogen and alterations occurring in the pathogen's genetic constitution before and after causing disease (Ai *et al.*, 2020). As there is no specific vaccine and medicine for COVID-19, it is necessary to diagnose them early to be isolated from healthy people. The latest instructions for diagnosing and treating COVID-19 pneumonia presented that the identification of COVID-19 should be confirmed by gene sequencing for blood and respiratory samples or by reverse transcription-polymerase chain reaction (RT-PCR), i.e., the critical markers for hospitalization (Tahamtan and Ardebili, 2020). WHO and the Centre for disease control (CDC) recognized that SARS-CoV-2 infection could be precisely identified by utilizing molecular methods on lower and upper respiratory tract pathogens. Therefore, the infection can be detected by utilizing real-time reverse-transcription polymerase chain reaction (rRT-PCR) tests, aiming at one or more genes in the SARS-CoV-2 genome (Lu *et al.*, 2020).

Nucleic acid testing

It is the leading approach to COVID-19 detection. Several RT-PCR kits have been produced for the genetic identification of SARS-CoV-2. It is a test involving reverse transcription of coronavirus with a DNA strand called complementary DNA (cDNA). Subsequently, various portions of cDNA are amplified or magnified (Wu *et al.*, 2020).

Sampling and viral load

A significant viral load in the lower and upper respiratory tract of COVID-19 patients has been observed within five to six days of symptoms appearing. Initial stage infection is usually analyzed and identified by using an oropharyngeal (OP) swab accompanied by a nasopharyngeal (NP) swab (Chen *et al.*, 2020). After collection, the sample is sent to the laboratory using viral transport, ideally in refrigerated conditions. It is worth considering that the virus may not get spotted by OP/NP swabs in the initial stage. In later infectious stages, the viral replication may get shifted to the patient's lower respiratory tract. In such a case, the sample should be taken from the lower respiratory tract (Xia *et al.*, 2020). The upper respiratory tract specimens are usually recommended, but a patient with a productive cough should take specimens from the lower respiratory tract. For upper respiratory tract sample collection, OP and NP swabs, nasal aspirates, and NP washes are utilized.

On the other hand, the lower respiratory tract specimen is collected from the sputum by using tracheal aspirates and bronchoalveolar lavage (BAL) fluid. The tracheal aspirates and BAL result in the formation of an aerosol. Viral load is based on the number of days spent before the

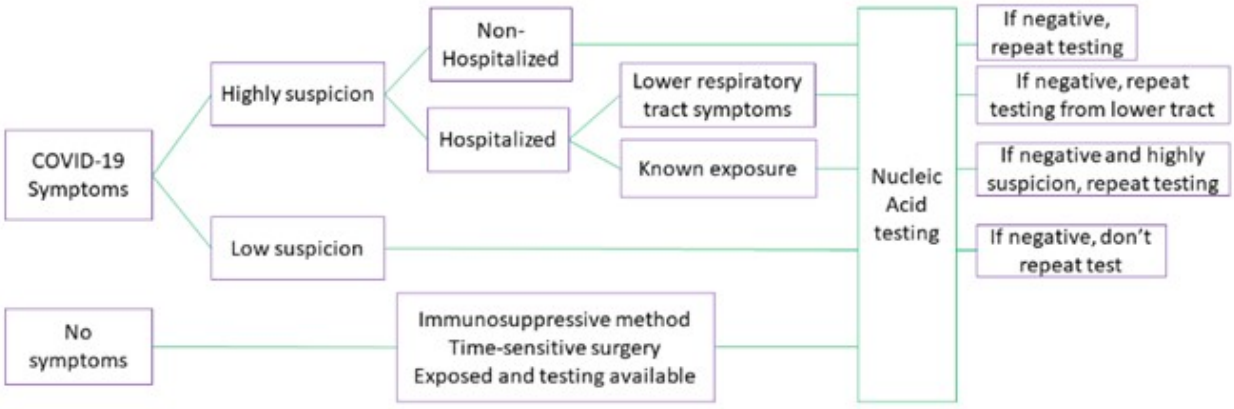

Fig. 2: Workflow for the symptomatic and asymptomatic patients of COVID-19.

occurrence of symptoms. The throat swabs become doubtful eight days after symptoms, whereas sputum specimens and NP swabs can be used 14 days after symptoms (Voiriot *et al.*, 2020). However, high viral RNA loads of SARS-CoV-2 are observed in the fecal matter of patients having COVID-19 pneumonia (Munster *et al.*, 2020).

Sample handling

SARS-CoV-2 is a single-stranded RNA with careful specimen handling and worthy laboratory instructions to isolate its genetic material. Before RNA extraction, several Chinese laboratories deactivate the virus, due to its contagious nature, by heating at 56-60C for about 30-60 min (Zhu *et al.*, 2020). The United States CDC employs a single-stage rRT-PCR test that provides quantitative data of viral loads for coronavirus detection.

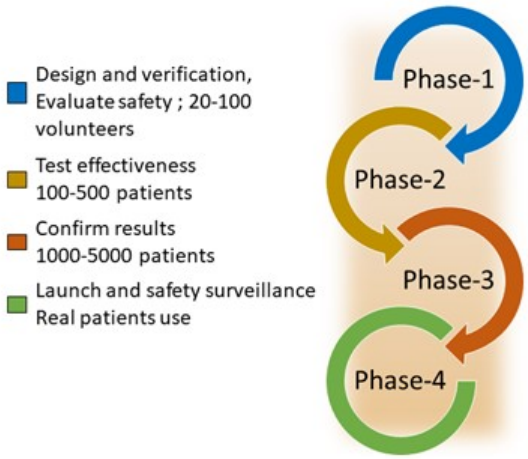

Fig. 3: Developmental phases of diagnostic tests. Phases 1 and 2 typically occur in an academic setting while phases 3 and 4 occur in a company after commercial transfer. Most diagnostic technologies are at the proof-of-concept stage and few are in phase 3 that can be quickly adapted for diagnosing pathogens in new outbreaks. The advancement of more phase 2 technologies into phase 3 would increase the number of approaches for detecting new pathogens.

Workflow of SARS-CoV-2

To execute the assay, viral RNA is isolated and then added to the master mix. The master mix comprises a fluorophore quencher probe, forward and reverses primers, nuclease-free water, reaction mix (magnesium, polymerase, nucleotides, reverse transcriptase, and excipients). The isolated RNA and the master mix are then put in the PCR thermocycler, and the incubation temperature is adjusted to perform the test. During rRT-PCR, a luminous pointer is formed by cutting the fluorophore quencher probe. The thermocycler detected the pointer, and then the magnified growth is recorded in real-time (A. Wu *et al.*, 2020). Corman *et al*. proposed a three-stage method for SARS-CoV-2 detection. These stages were classified as first-line screening, verification, and discriminatory tests. The first line screening detects all the viruses related to SARS by targeting different viral gene sites. If the result is positive, the detection of the RdRP gene was recommended, for which two probes and two primers are employed. A discriminatory assay is executed in a positive test using one of the two probe sequences (Sexton *et al.*, 2016).

Computed tomography

CT scan of the chest is a method comprising X-rays proportions at various directions on the patient's chest to form cross-sectional images. Radiologists analyze the strange characters in the images that lead to detection (Tang *et al.*, 2020). CT scanner creates various estimations from different rotational angles of X-ray attenuation by the thorax's cross-sectional level. The statistics thus obtained are employed to form digital imaging, which depicts the thickness of the targeted section (Wrapp *et al.*, 2020).

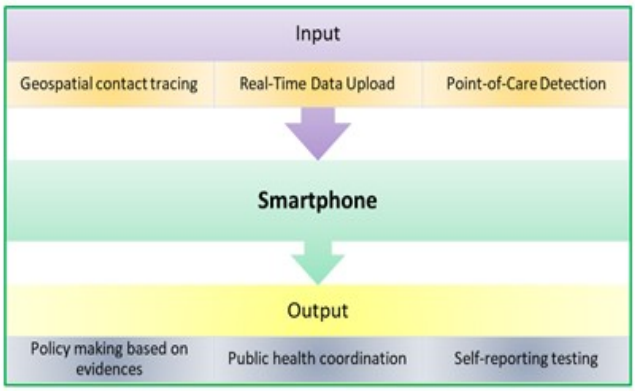

Fig. 4: Role of smartphones in diagnostics. Smartphone capabilities such as connectivity, databasing, and onboard hardware enable better evidence-based policy making, national disease response coordination and community healthcare.

Chest CT for detection of coronavirus

The low sensitivity of RT-PCR shows that various COVID-19 cases may not be recognized and may not get timely cure. Such patients enhance the chance of spreading the infection in various individuals, owing to the virus transmissible mode. The chest CT scan is relatively simple to achieve and can rapidly detect COVID-19 pneumonia. Thus, CT is beneficial for the identification of COVID-19. Typical radiographic characteristics such as interstitial changes with a peripheral distribution, multifocal patchy consolidation, and ground-glass opacities are displayed in nearly all patients (Lim *et al.*, 2016).

Imaging characteristics of COVID-19

The chest CT of COVID-19 patient has diverse imaging characteristics based on the infectious stage, followed by symptoms. Bernheim *et al*. observed that CT imaging exhibited clear and normal lungs in the initial stages of infection, but after ten days of symptoms occurrence, lung peaking was displayed. The typical features of COVID-19 are marginal and two-sided ground-glass opacities (hazy section in lungs) and fluid-filled compressible lung tissues, which are displayed as lung whitening in chest CT (Neuman *et al.*, 2011).

Serological immunoassay

Few serum assays for detecting COVID-19 have been formulated by the in-vitro diagnostic (IVD) companies. The serum assay is also called a serological immunoassay. This assay involves the detection of antibodies and viral proteins in the plasma and serum. SARS-CoV-2 infection is detected at the commercial level by employing biomarkers such as manual ELISA, automated chemiluminescence immunoassay (CLIA), and rapid lateral flow immunoassay (LFIA) assays. It adopts a principle that antibodies like IgG and IgM are formed from the second week of disease. IgG is detected in the patient sample from 20 days of infection and afterward, whereas IgM is detected during 10-30 days after infection (Schoeman and Fielding, 2019). RT-PCR based in-vitro diagnostic assays are utilized all over the world to confirm the diagnosis of COVID-19. By the second week of January 2020, Tib-Molbiol, Germany, in collaboration with several companions, developed a modern and more potent rRT-PCR.

Emerging assay for COVID-19

According to WHO, the prime and leading concern in the analytical study of COVID-19 is the development of protein and nucleic acid assays and a patient diagnosis at the bedside, which is commonly called point-of-care testing (POC). Remarkable advancement has been made in in-vitro diagnostic assays. rRT-PCR is the primary in-vitro diagnostic assay for COVID-19, though it requires hours to complete (Schoeman and Fielding, 2019).

Nucleic acid amplification

Nucleic acid testing (NAT) directly review the DNA sequencing, collecting correlated clinical data from either pathogen or patient. NAT is significant due to its capability to detect pathogenicity indicators, antibiotic tolerance, and identifying immensely precise genotyping, thus permitting timely treatment and therapeutic intervention (Hamming *et al.*, 2004).

Loop-mediated isothermal amplification (LAMP)

Nowadays, efforts are being made to advance NAT by employing isothermal amplification for SARS-CoV-2 identification. Isothermal amplification is a single tube method performed at a fixed temperature, and no particular laboratory instrument is needed to create similar analytical reactivity to a polymerase chain reaction (Holshue *et al.*, 2020). LAMP is known to be a particular reaction comprising 60-65C temperature and a minimum of 4 primers. It is based on the principle that amplification leads to detection. Analytical reactivity has been proven to surpass that of equally efficient PCR assays with diagnostic limits as low as five copies (Zou *et al.*, 2020).

Reverse transcription LAMP comprises DNA polymerase and about 4-6 primers, which binds to 6 distinct positions on the targeted genetic material. In 4 primers, two primers are considered an outer primer, and the other two are inner primers. LAMP employs a large number of primers relative to other methods, leading to precise results. During LAMP analytical assay, the specimen is placed in the tube, and the amplified DNA is detected by the color appearing on the incorporation of pH-sensitive dye, turbidity occurring as a reaction by-product, and fluorescence appearing on the introduction of a fluorescent dye (bind to double-stranded DNA) (Guan *et al.*, 2020).

Point of care (POC) testing

The introduction of PCR advanced genetics and molecular detection results in a well-designed nucleic acid amplification technique by employing thermostable polymerase enzymes, repetitive heating, and cooling to achieve strand extraction and annealing. The heating cycle demands a complex strategy along with a constant power source for PCR execution, thus forming micro-scale PCR an unachievable solution for inclusion into POC programs. Whereas, confining the PCR reaction to the microfluidic system demands additional examinations like thermal restrictions of substance and the complex heat control system, excessive water evaporation, and heat sensitivity of enzymes being employed in detection (Sheridan, 2020).

POC assay is executed to identify patients without the need for dispatching samples at various laboratories; thus, the communities lacking centralized laboratories can also diagnose the infected patient. Lateral flow antigen detection is among the under-developed POC assay for SARS-CoV-2 (Wu *et al.*, 2020). The development phases of diagnostic tests are summarized in fig. 3.

Point-of-care utilizing LAMP

Current diagnostic methods are time taking and demand skilled workers for performing tasks, so there is a need to create a substitutive amplification method. LAMP is an extremely precise, speedy, and efficient method that amplifies DNA in isothermal settings. This method employs four specially designed primers as well as DNA polymerase, which plays a role in stand displacement (Miller *et al.*, 2020). While executing the LAMP, instead of denaturation from heat, the DNA polymerase is utilized to form a single-stranded design; thus, the task can be performed in an isothermal state. Furthermore, there is no difficulty left regarding power supply and a thermocycler. Hence, LAMP is regarded as relatively more stable. In the POC device, LAMP is utilized to detect zoonotic illness, which produces respiratory symptoms like the Avian influenza virus (AIV) in the VIVALDI (Veterinary validation of point-of-care diagnostic instrument) strategy. The IgG/IgM Rapid Test's sensitivity is about 88.66%, which is thought to be lesser than assays' reactivity based on LAMP-reaction (greater than 95%).

Song's group created the penn-RAMP to raise the sensitivity of COVID-19 identification assays. penn-RAMP is a two-step isothermal dsDNA amplification method that merges LAMP and recombinase polymerase amplification (RPA) methods in one tube. For performing LAMP assay, Song *et al*. selected preserved sequences of COVID-19 by employing Clustal X and LAMP primers. To simplify the method, leucocrystal violet (LCV) dye was utilized as a coloring agent, imparted deep violet color visible with the naked eye. This detection method was relatively easy, and just one tube was required to carry out the procedure (Goodwin *et al.,* 2016).

Point-of-care with microfluidic devices

POC analysis can be done by utilizing microfluidic devices. The device comprises a chip of size almost equal to palm, carved with reaction chambers and micrometer-sized channel. The chips are composed of paper, glass, or poly-dimethylsulfoxide. These chips combine and segregate the liquid specimens by utilizing various forces like capillary, electrokinetic, or vacuum. There are several benefits of using microfluidic devices: speedy analysis, simple movability, miniaturization, and a small specimen. These devices are employed to identify the proteins and RNA of coronavirus (Freeman, Walker and Vrana, 1999).

Smartphone Surveillance

Management of epidemics demands immense observation, interchanging of epidemiological information, and patient examination (Gates, 2020). Healthcare systems need devices that can play a role in making communication easy and fast, to control the spreading of infection. A smartphone is highly useful as it has computational power, an epidemiological database, hardware to assist electronic reporting, connectivity, and POC analysis (fig. 4) (Nayak *et al.*, 2017). The increase in smartphone usage worldwide makes it a convenient tool to coordinate responses during COVID-19 (Wood *et al.*, 2019). Inadequate dealing, as well as under-reporting, escalates the worldwide spreading of COVID-19. For instance, Iran reported the first 43 cases on February 23 2020, the death rate was around 19%, and three patients moved abroad from Iran. After considering this report, transmission modeling presented that the cases of COVID-19 in Iran were in thousands (Tuite *et al.*, 2020).

Smartphones can link with analytical assays to give real-time geospatial statistics, which authorizes worldwide healthcare organizations to execute coordinated control plans. Various research associations have utilized smartphones for the geospatial tracing of infections like Ebola, HIV, and TB (Brangel *et al.*, 2018; Danquah *et al.*, 2019). During the time of Ebola, smartphones were employed for contact tracking, a method used for analyzing and tracing people who were in contact with any Ebola patient and may get infected (Danquah *et al.*, 2019). Smartphones can digitize contact tracking in order to offer shareable data.

Lack of contact between regional healthcare associations leads to varying transmission rates worldwide (Skowronski *et al.*, 2006) like in Canada in 2003 during the SARS epidemic. Around 247 cases were reported by Toronto, Ontario, out of which 3 cases were imported. On the other hand, just 5 cases were reported by Vancouver, British Columbia, 3 out of 5 cases were imported (Skowronski *et al.*, 2005). Ontario had no provincial public health agency at that time, whereas Columbia's agency had already detected that the province was a significant threat of importing the infectious outbreak.

Before the SARS epidemic, Columbia's agency developed a digital network to ease the province's correspondence. These digital networks can broaden by using smartphone connectivity. The smartphones are employed to post and share the information of the outbreak on the communal database and coordinate epidemiological feedback.

COVID-19 suspected patients may face difficulty in contacting their clinicians. People showing light respiratory signs may feel reluctant to come to the hospitals because they experience a significant threat of having COVID-19. The usage of smartphones offers direct contact of the patient with the health care professional without getting the disease. During the 2009 influenza epidemic, Switzerland employed medical teleconsultations to control the expected cases besides the present reporting setup (Blozik, Grandchamp and von Overbeck, 2012). Relative to the in-person discussion, teleconsultation results in increased influenza reporting because of fewer complications in access. When a COVID-19 patient is analyzed in the hospital and the test results positive, then the patient with mild symptoms is recommended to go home and self-quarantine. This self-quarantine lessens the contact between the patient and health care professional; thus, the clinician's patient examination decreases, causing unfavorable effects on the patient's mental health. The smartphone applications connect the patient with the counselor to face the loneliness and distress in quarantine and disease (Xiang *et al.*, 2020).

Moreover, it enables the patient to describe his symptoms and feelings, which assists the clinician's distant patient examination (Karimuribo *et al.*, 2017). Smartphone-based reporting is useful in giving epidemiologists the data related to excessive spreading methods of disease. For instance, in the MERS 2013 epidemic, passengers were monitored during the Hajj pilgrimage by utilizing smartphone applications. In this smartphone application, the users informed the beginning of symptoms, exposure to animals, and hand hygiene protocols in pilgrimage and after going back to their own countries (Alqahtani *et al.*, 2016). Many other applications can be utilized to provide information to the health agencies and upgrade feedback to infections outbreak.

Currently, remarkable success has been made in combining analytical technology and smartphone. Instead of using typical laboratory instruments, the modules of smartphone-like auto jack, flashlight and camera are employed for interpreting the identification tests (Malekjahani *et al.*, 2019). Thus, the usage of these devices makes the diagnostic workflow simplified by automating interpretation and database. For instance, the analysis of a smartphone-based microscope in Cameroon revealed a rapid turnaround time relative to standard methods (D'Ambrosio *et al.*, 2015). Kanazawa *et al.* confirmed that smartphones with FLIR (Forward Looking Infrared Radar) could be used in analyzing the body temperature in case of inflammation. The device can also be used in diagnosing fever, which is a typical symptom of several SARS-CoV-2 infections (Kanazawa *et al.*, 2016). A smartphone-based microscope created by Mudanyali *et al*. transfers diagnostic outcomes to the database for examination as well as Spatio-temporal mapping (Wu and McGoogan, 2020). This technology deals with the requirement of POC assay at a collaborative platform.

Concluding remarks and viewpoint

The design of COVID-19 detection and tracking has been motivated by lessons learned from the 2002 SARS epidemic. Diagnostics are a vital aspect of the epidemic managing tools since they help medical professionals to guide patients to services and initiatives. Serological and point-of-care assessments are now the phase in the method of diagnosis. More contact and monitoring, claim scientists, could be enabled by the mixture of diagnostic techniques and smartphones. For timely detection or monitoring for SARS-CoV-2, a nasopharyngeal instead of an oropharyngeal swab is suggested. The function of rectal swabs in evaluating late illness patients or as an infectivity/treatment measure is not presently extensively known. It is essential to consider the value of regular tests or bronchoscopy usage in people with the critical disease even though the first diagnostic test is negative. Dr. Richard Schmitz said that it is difficult to overstate the exponential advancement of interconnected, random-access, point-of-care molecular instruments for effective diagnosis. He said these tests are reliable, easy, and quick and can be used in community medical clinics.

Smartphone-based sensors have started to gain significant concerns as they provide a semi-automated user experience that they can use without formal training or specialized experienced by ordinary people. With inbuilt customized software and signaling, tools can be prepared inside a smartphone to wirelessly transmit the process to be worked out at any place and can be controlled by semi-trained workers. A cost-effective solution for costly stand-alone technology could be offered by smartphone-driven monitoring and diagnostic devices. A COVID-19 biosensor can be built by incorporating a complete disposable sensing module into a smartphone-based application framework for personalized diagnosis, as discussed before. The sensing surface can be optimized according to the marker molecules and can be improvised further in real clinical samples for POC diagnosis. In addition to detecting and tracking the outbreak on a broad scale, such a miniaturized device can provide rapid and inexpensive sensing.

REFERENCES

Ai T, Yang Z, Hou H, Zhan C, Chen C, Lv W, Tao Q, Sun Z and Xia L (2020). Correlation of chest CT and RT-PCR testing in coronavirus disease 2019 (COVID-19) in China: A report of 1014 cases, *Radiology,* **296**(2): 200642.

Alqahtani AS, BinDhim NF, Tashani M, Willaby HW, Wiley KE, Heywood AE, Booy R and Rashid H (2016). Pilot use of a novel smartphone application to track traveller health behaviour and collect infectious disease data during a mass gathering: Hajj Pilgrimage 2014. *J, Epidemiol Glob Health*, **6**(3): 147-155.

Blozik E, Grandchamp C and von Overbeck J (2012). Influenza surveillance using data from a telemedicine centre. *Int. J. Public Health*, **57**(2): 447-452.

Brangel P, Sobarzo A, Parolo C, Miller BS, Howes PD, Gelkop S, Lutwama JJ, Dye JM, McKendry RA, Lobel L and Stevens MM. (2018). A serological point-of-care test for the detection of IgG antibodies against Ebola virus in human survivors. *ACS Nano,* **12**(1): 63-73.

Chauhan DS, Prasad R, Srivastava R, Jaggi M, Chauhan SC and Yallapu MM. (2020). Comprehensive Review on Current Interventions, Diagnostics and Nanotechnology Perspectives against SARS-CoV-2. *Bioconjugate Chem,* **31**(9): 2021-2045.

Chen J, Liu D, Liu L, Liu P, Xu Q, Xia L, Ling Y, Huang D, Song S, Zhang D and Qian Z (2020). A pilot study of hydroxychloroquine in treatment of patients with common coronavirus disease-19 (COVID-19). *J. Zhejiang Univ. (Med. Sci)*: **49**(2): 215-219

D'Ambrosio MV, Bakalar M, Bennuru S, Reber C, Skandarajah A, Nilsson L, Switz N, Kamgno J, Pion S, Boussinesq M and Nutman TB (2015). Point-of-care quantification of blood-borne filarial parasites with a mobile phone microscope. *Sci Transl Med.,* **7**(286): 286re4-286re4.

Danquah LO, Hasham N, MacFarlane M, Conteh FE, Momoh F, Tedesco AA, Jambai A, Ross DA and Weiss HA (2019). Use of a mobile application for Ebola contact tracing and monitoring in northern Sierra Leone: a proof-of-concept study. *BMC Infect Dis.,* **19**(1): 810.

Van Doremalen N, Bushmaker T, Morris DH, Holbrook MG, Gamble A, Williamson BN, Tamin A, Harcourt JL, Thornburg NJ, Gerber SI and Lloyd-Smith JO (2020). Aerosol and surface stability of SARS-CoV-2 as compared with SARS-CoV-1. *N Engl J Med.,* **382**(16): 1564-1567.

Freeman WM, Walker SJ and Vrana KE. (1999). Quantitative RT-PCR: Pitfalls and potential. *Biotech.,* **26**(1): 112-125.

Fu L, Wang B, Yuan T, Chen X, Ao Y, Fitzpatrick T, Li P, Zhou Y, Lin YF, Duan Q and Luo G. (2020). Clinical characteristics of coronavirus disease 2019 (COVID-19) in China: A systematic review and meta-analysis. *J. Infect.,* **80**(6): 656-665.

Gates B (2020). Responding to Covid-19-a once-in-a-century pandemic? *N. Engl. J. Med.,* **382**(18): 1677-1679.

Di Gennaro F, Pizzol D, Marotta C, Antunes M, Racalbuto V, Veronese N and Smith L. (2020). Coronavirus diseases (COVID-19) current status and future perspectives: A narrative review. *Int. J. Environ. Res. Public Health,* **17**(8): 2690.

Goodwin S, McPherson JD and McCombie WR (2016). Coming of age: Ten years of next-generation sequencing technologies. *Nat. Rev. Genet.,* **17**(6): 333.

Guan WJ, Ni ZY, Hu Y, Liang WH, Ou CQ, He JX, Liu L, Shan H, Lei CL, Hui DS and Du B (2020). Clinical characteristics of coronavirus disease 2019 in China. *N. Engl. J. Med.,* **382**(18): 1708-1720.

Hamid H, Masood RA, Tariq H, Khalid W, Rashid MA and Munir MU. (2020). Current pharmacy practices in low-and middle-income countries; Recommendations in response to the COVID-19 pandemic. *Drugs Ther. Perspect*., **36**: 355-357.

Hamid H, Masood RA, Khalid W, Saqlain M, Tariq H and Munir MU (2020). Emerging pharmacy services: Recommendations for emergency care of COVID-19 pandemic in low and middle-income countries. *Pak. J. Pharm. Sci*, **33**(4): 1735-1738.

Hamming I, Timens W, Bulthuis ML, Lely AT, Navis GV and van Goor H (2004). Tissue distribution of ACE2 protein, the functional receptor for SARS coronavirus. A first step in understanding SARS pathogenesis. *J. Pathol. Soc. Great Britain and Ireland,* **203**(2): 631-637.

Holshue ML, DeBolt C, Lindquist S, Lofy KH, Wiesman J, Bruce H, Spitters C, Ericson K, Wilkerson S, Tural A and Diaz G (2020). First case of 2019 novel coronavirus in the United States. *N. Engl. J. Med.,* **382**: 929-936.

Ji W, Wang W, Zhao X, Zai J and Li X (2020). Cross-species transmission of the newly identified coronavirus 2019-nCoV. *J Med. Virol.,* **92**(4): 433-440.

Joshi RS, Jagdale SS, Bansode SB, Shankar SS, Tellis MB, Pandya VK, Chugh A, Giri AP and Kulkarni MJ (2020). Discovery of potential multi-target-directed ligands by targeting host-specific SARS-CoV-2 structurally conserved main protease. *J. Biomol. Struct. Dyn*., pp.1-16.

Kanazawa T, Nakagami G, Goto T, Noguchi H, Oe M, Miyagaki T, Hayashi A, Sasaki S and Sanada H (2016). Use of smartphone attached mobile thermography assessing subclinical inflammation: A pilot study. *J. Wound Care,* **25**(4): 177-182.

Karimuribo ED, Mutagahywa E, Sindato C, Mboera L, Mwabukusi M, Njenga MK, Teesdale S, Olsen J and Rweyemamu M. (2017). A smartphone app (AfyaData) for innovative one health disease surveillance from community to national levels in Africa: intervention in disease surveillance. *JMIR Public Health Surveill.,* **3**(4): e94.

Kobayashi T, Jung SM, Linton NM, Kinoshita R, Hayashi K, Miyama T, Anzai A, Yang Y, Yuan B, Akhmetzhanov AR and Suzuki A. (2020). Communicating the risk of death from novel coronavirus disease (COVID-19). *J. Cli.n Med.,* **9**(2): 580.

Li X, Zai J, Wang X and Li Y (2020). Potential of large "first generation" human-to-human transmission of 2019-nCoV. *J. Med. Virol.,* **92**(4): 448-454.

Lim YX, Ng YL, Tam JP and Liu DX (2016). Human coronaviruses: A review of virus–host interactions. *Diseases,* **4**(3): 26.

Liu C, Zhou Q, Li Y, Garner LV, Watkins SP, Carter LJ, Smoot J, Gregg AC, Daniels AD, Jervey S and Albaiu D (2020). Research and development on therapeutic agents and vaccines for COVID-19 and related human coronavirus diseases. *ACS Cent. Sci.,* pp.315-331.

Lu R, Zhao X, Li J, Niu P, Yang B, Wu H, Wang W, Song H, Huang B, Zhu N and Bi Y (2020). Genomic characterisation and epidemiology of 2019 novel coronavirus: Implications for virus origins and receptor binding. *The Lancet.,* **395**(10224): 565-574.

Lu X, Wang L, Sakthivel SK, Whitaker B, Murray J, Kamili S, Lynch B, Malapati L, Burke SA, Harcourt J and Tamin A

(2020). US CDC real-time reverse transcription PCR panel for detection of severe acute respiratory syndrome coronavirus 2, *Emerg. Infect. Dis*., **26**(8): 1654.

Malekjahani A, Sindhwani S, Syed AM and Chan WC. (2019). Engineering Steps for Mobile Point-of-Care Diagnostic Devices. *Acc Chem Res.,* **52**(9): 2406-2414.

Miller S, Chiu C, Rodino KG and Miller MB. (2020). Point-Counterpoint: Should We Be Performing Metagenomic Next-Generation Sequencing for Infectious Disease Diagnosis in the Clinical Laboratory? *J Clin Microbiol.,* **58**(3): e01739-19.

Mizumoto K, Kagaya K, Zarebski A and Chowell G. (2020). Estimating the asymptomatic proportion of coronavirus disease 2019 (COVID-19) cases on board the Diamond Princess cruise ship, Yokohama, Japan. *Eurosurveillance.,* **25**(10): 2000180.

Munster VJ, Feldmann F, Williamson BN, van Doremalen N, Pérez-Pérez L, Schulz J, Meade-White K, Okumura A, Callison J, Brumbaugh B and Avanzato VA. (2020). Respiratory disease in rhesus macaques inoculated with SARS-CoV-2. *Nature.,* **585**(7824): 268-272.

Nayak S, Blumenfeld NR, Laksanasopin T and Sia SK. (2017). Point-of-care diagnostics: Recent developments in a connected age. *Anal Chem.,* **89**(1): 102-123.

Neuman BW, Kiss G, Kunding AH, Bhella D, Baksh MF, Connelly S, Droese B, Klaus JP, Makino S, Sawicki SG and Siddell SG. (2011). A structural analysis of M protein in coronavirus assembly and morphology. *J Struct Biol.,* **174**(1): 11-22.

Oran DP and Topol EJ (2020). Prevalence of Asymptomatic SARS-CoV-2 Infection: A Narrative Review. *Ann Inter Med.,* **173**(5): 362-367.

Schoeman D and Fielding BC (2019) 'Coronavirus envelope protein: current knowledge', *Virol J.,* **16**(1): 1-22.

Sexton NR, Smith EC, Blanc H, Vignuzzi M, Peersen OB and Denison MR. (2016). Homology-based identification of a mutation in the coronavirus RNA-dependent RNA polymerase that confers resistance to multiple mutagens. *J Virol.,* **90**(16): 7415-7428.

Sheridan C (2020). Coronavirus and the race to distribute reliable diagnostics. *Nat Biotechnol.*, **38**(4): 382.

Skowronski DM, Astell C, Brunham RC, Low DE, Petric M, Roper RL, Talbot PJ, Tam T and Babiuk L. (2005). Severe acute respiratory syndrome (SARS): A year in review. *Annu. Rev. Med. Ann Rev.,* 56(18): 357-381.

Skowronski DM, Petric M, Daly P, Parker RA, Bryce E, Doyle PW, Noble MA, Roscoe DL, Tomblin J, Yang TC and Krajden M. (2006). Coordinated response to SARS, Vancouver, Canada. *Emerg Infect Dis*., **12**(1): 155.

Tahamtan A and Ardebili A (2020). Real-time RT-PCR in COVID-19 detection: Issues affecting the results. *Expert Rev Mol Diagn.* 20(5): 453-454.

Tang X, Wu C, Li X, Song Y, Yao X, Wu X, Duan Y, Zhang H, Wang Y, Qian Z and Cui J (2020). On the origin and continuing evolution of SARS-CoV-2. *Natl Sci Rev*. 7(6): 1012-1023.

Tuite A, Ng V, Rees E and Fisman D. (2020). Estimation of COVID-2019 burden and potential for international dissemination of infection from Iran. *Med. Rxiv.* **20**(8): 894.

Verma N, Patel D and Pandya A. (2020). Emerging diagnostic tools for detection of COVID-19 and perspective. *Biomed microdevices.,* **22**(4): 1-18.

Voiriot G *et al.* (2020). Bronchoalveolar lavage findings in severe COVID-19 pneumonia. *Intern Emerg Med.,* 1333-1334.

Voiriot G, Fajac A, Lopinto J, Labbé V and Fartoukh M. (2020). Unique epidemiological and clinical features of the emerging 2019 novel coronavirus pneumonia (COVID-19) implicate special control measures. *J. Med. Virol.,* **92**(6): 568-576.

Wood CS, Thomas MR, Budd J, Mashamba-Thompson TP, Herbst K, Pillay D, Peeling RW, Johnson AM, McKendry RA and Stevens MM. (2019). Taking connected mobile-health diagnostics of infectious diseases to the field. *Nature,* **566**(7745): 467-474.

Wrapp D, Wang N, Corbett KS, Goldsmith JA, Hsieh CL, Abiona O, Graham BS and McLellan JS. (2020). Cryo-EM structure of the 2019-nCoV spike in the prefusion conformation. *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 and Sheng J. (2020). Genome composition and divergence of the novel coronavirus (2019-nCoV) originating in China. *Cell Host Microbe.,* **27**(3): 325-328.

Wu F, Zhao S, Yu B, Chen YM, Wang W, Song ZG, Hu Y, Tao ZW, Tian JH, Pei YY and Yuan ML. (2020). A new coronavirus associated with human respiratory disease in China. *Nature.,* **579**(7798): 265-269.

Wu J, Liu J, Li S, Peng Z, Xiao Z, Wang X, Yan R and Luo J (2020). Detection and analysis of nucleic acid in various biological samples of COVID-19 patients. *Travel. Med. Infect. Dis.,* 37: 101673.

Wu Z and McGoogan JM (2020). Characteristics of and important lessons from the coronavirus disease 2019 (COVID-19) outbreak in China: Summary of a report of 72 314 cases from the Chinese Center for Disease Control and Prevention. *Jama. American Medical Association,* **323**(13): 1239-1242.

Xia W, Shao J, Guo Y, Peng X, Li Z and Hu D. (2020) Clinical and CT features in pediatric patients with COVID-19 infection: Different points from adults, *Pediatric pulmonology.,* **55**(5): 1169-1174.

Xiang YT, Yang Y, Li W, Zhang L, Zhang Q, Cheung T and Ng CH (2020). Timely mental health care for the 2019 novel coronavirus outbreak is urgently needed. *The Lancet Psychiatry.,* **7**(3): 228-229.

Zhou P, Yang XL, Wang XG, Hu B, Zhang L, Zhang W, Si HR, Zhu Y, Li B, Huang CL and Chen HD (2020). A pneumonia outbreak associated with a new coronavirus of probable bat origin. *Nature,* **579**(7798): 270-273.

Zhu N, Zhang D, Wang W, Li X, Yang B, Song J, Zhao X, Huang B, Shi W, Lu R and Niu P (2020). A novel coronavirus from patients with pneumonia in China, 2019. *N. Engl. J. Med.,* **382**: 727-733.

Zou L, Ruan F, Huang M, Liang L, Huang H, Hong Z, Yu J, Kang M, Song Y, Xia J and Guo Q (2020). SARS-CoV-2 viral load in upper respiratory specimens of infected patients. *N. Engl. J. Med.,* **382**(12): 1177-1179.