

Pandemic SARS Coronavirus-2 Infections in Humans- COVID-19

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Abstract

The Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) first broke out in Wuhan (China) and subsequently spread worldwide. Coronaviruses (CoVs) primarily cause zoonotic infections in birds and mammals however, in the last few decades have shown to be capable of infecting humans as well. The outbreak of severe acute respiratory syndrome (SARS) in 2003 and more recently, Middle-East respiratory syndrome, (MERS) has demonstrated the lethality of CoVs when they cross the species barrier and infect humans. Coronavirus (CoV) is a large family of viruses that cause afflictions ranging from the common cold to more severe pathologies such as Middle East Respiratory Syndrome (MERS-CoV) and Severe Acute Respiratory Syndrome (SARS-CoV). A novel coronavirus (nCoV) is a new strain that has now been identified in humans. The recognition of a new coronavirus identified in December 2019, named CoVID-19 are common for coronavirus researchers. Detailed investigations found that SARS Coronavirus-2 was initially transmitted from civets to humans and MERS-CoV from dromedary camels to humans. Advances in biology have resulted in a greater understanding of coronavirus, including them to adapt to new environments, trans-species infection and the emergence of new subtypes. New tools of cell and molecular biology have led to an increased understanding of intracellular replication and viral cell biology. Along with the advent of reverse genetic approaches in the past five years; it is now possible to begin to define the

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determinants of viral replication, trans-species adaptation, and human disease. The most progress has been made on SARS-CoV 2, highlighting specific structural requirements for its functions in the CoV life cycle as well as mechanisms behind its pathogenesis. In this review, we will provide a through insight to the life cycle of CoV, its genetics, replication process and reverse genetic applications to SCoV along with advances in its research. This review aims to establish the current knowledge on CoV-2 by highlighting the recent progress that has been made and comparing it to previous knowledge. We also conclude with a brief discussion on practices to decrease risk factors for transmission and treatment options.

Keywords: Human coronavirus, SARS coronavirus-2, infections in humans-COVID-19.

İnsan-COVID-19'da Pandemik SARS Coronavirus-2 Enfeksiyonları

Öz

İlk olarak Wuhan'da (Çin) patlak veren Şiddetli Akut Solunum Sendromu Koronavirüs 2 (SARS-CoV-2), sonrasında dünyaya yayılmıştır. Koronavirüsler (CoVs) başlarda kuşlarda ve memelilerde zoonotik enfeksiyonlara neden olurken, son birkaç yılda insanları da enfekte edebildiği gösterilmiştir. 2003 yılında Şiddetli Akut Solunum Sendromu'nun (SARS) ve son zamanlarda Orta Doğu Solunum Sendromunun (MERS) patlak vermesi, CoV'lerin tür bariyerini geçip insanları enfekte ettiklerinde ölümcül olduklarını göstermiştir. Koronavirüs (CoV), soğuk algınlığından Orta Doğu Solunum Sendromu (MERS-CoV) ve Şiddetli Akut Solunum Sendromu (SARS-CoV) gibi daha şiddetli patolojilere kadar çeşitli rahatsızlıklara neden olan büyük bir virüs ailesidir. Yeni koronavirüs (nCoV), insanlarda daha önce tanımlanmamış yeni bir suş türüdür. Aralık 2019'da tanımlanan yeni koronavirüs, araştırmacılar tarafından genel olarak CoVID-19 olarak adlandırılmıştır. Yapılan ayrıntılı araştırmalarda, SARS Coronavirus-2'nin başlangıçta misk kedilerinden, MERS-CoV'un ise tek hörgüçlü develerden insanlara bulaştığı bulunmuştur. Biyoloji alanındaki gelişmeler koronavirüsün, yeni ortamlara adaptasyonu, trans-tür enfeksiyonu ve yeni alt tiplerinin ortaya çıkması da dahil olmak üzere daha iyi anlaşılmasını sağlamıştır. Hücre ve moleküler biyolojide kullanılan yeni araçlar, hücre içi replikasyon ve viral hücre biyolojisinin daha iyi anlaşılmasını sağlamıştır. Son beş yılda ters genetik yaklaşımların ortaya çıkmasıyla birlikte; artık viral replikasyonun, trans-tür adaptasyonunun ve insan hastalığının belirleyicilerini tanımlamaya başlamak mümkündür. Bu derlemede, CoV'nin yaşam döngüsü, genetiği, replikasyon süreci ve SCoV'ye ters genetik uygulamalarının araştırılması ile ilgili ilerlemelerle ilgili bilgi verilecektir. Bu derleme, yakın zamanda kaydedilen ilerlemeyi vurgulayarak ve önceki bilgilerle karşılaştırarak CoV-2 hakkındaki mevcut bilgileri oluşturmayı amaçlamaktadır. Ayrıca, bulaşma ve tedavi seçenekleri için risk faktörlerini azaltmaya yönelik uygulamalar hakkında kısa bir tartışma ele alınmıştır.

Anahtar Kelimeler: İnsan koronavirüsü, SARS koronavirüs-2, insan-COVID-19 enfeksiyonları.

Introduction

Life Cycle of Coronavirus

Advances in biology have resulted in a greater understanding of coronavirus, including structure genome and nucleic acid sequence. The Coronavirus genome is a linear RNA particle consisting of a spike, an enveloped layer and a nucleic acid sequence with a single strand of mRNA¹. The genome is composed of the nucleocapsid protein N and contain an RNA nucleocapsid helical structure. The S protein is the evidence of pathogenesis as observed in mice. It contains species specificity, range, cell tropism and a disease marker². On the host cell area, virus replication initiates when the S protein conforms to particular receptors. Morphologically the genome of coronaviruses is single-stranded, linear mRNA molecules with polarity and diversity of lengths ranging from 28 to 32 kb³. Aminopeptidase N is the receptor in human coronavirus HCoV-229E. The precise mechanisms of entry and uncoating have not been defined, but likely occur by either fusion from viral and host cell membrane or through endocytic vesicles. The next discrete stage in the life cycle is translation and proteolytic processing of viral replicase proteins from the input genome RNA, followed by formation of cytoplasmic replication complexes in association with cellular membranes⁴. Mechanisms by which replication products are delivered to sites of assembly remain to be determined. It has been shown that subpopulations of replicase proteins and the structural nucleocapsid (N), translocate from replication complexes to sites of assembly and may mediate the process in association with cellular membrane protein pathways⁵. The endoplasmic reticulum Golgi intermediate compartment (ERGIC) viral congregation occurs both rapidly and physically distinct from viral replication structures. The replication mechanisms involved in the sites of influence, remain to be determined. It has been observed that replication protein and nucleocapsid N, alter from replication organization place together and arbitrate this system in combination with cell protein film pathways⁶. The genome RNA N, layer protein E and protein M adhere into the lumen of the ER/Golgi virosomes, which results in the budding and formation of particles⁷.

Viral Replication

The 5' replicase superiority of the RNA genome divides into co-amino terminal replicase polyproteins. For a continuous reproductive life cycle; coronavirus infection replication needs constant replicase quality of translation⁸. Replicase proteins arbitrate through positive and negative strand, genomic and subgenomic RNA production, as a result of capping, polyadenylation, RNA recoiling, arrangement change during viral RNA processing, and alternating transcription and transcription attenuation⁹. With the advent of SARS, more extensive bioinformatics analyses have resulted in predictions of several additional functions involved in RNA processing, including methyltransferase and exonuclease activities¹⁰.

Replicase Protein Expression

Coronaviruses activity incorporate both picornavirus 3C-like proteinase and papain-like proteinase (PLP) which are needed for encoding inside the replicase polyproteins, which equally mediate both trans and cis cleavage trial¹¹. There are sites for replicase polyproteins by amino acid sequences and the region of the replicase which experience the majority variation, suggestive for encoding and adornment functions or specific host jobs which are responsible to demands for quick additional alteration¹².

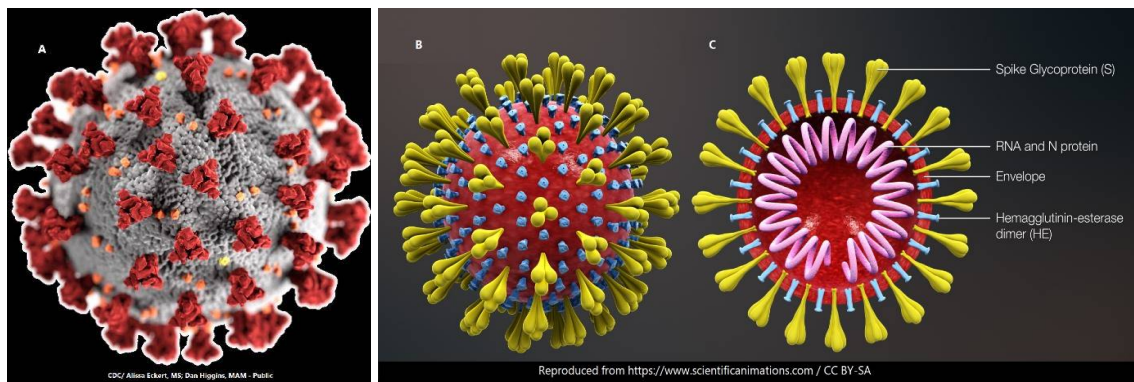


Figure 1: (A) Ultrastructural morphology exhibited by coronaviruses.

Figure 1: (B) 3D medical Images, depicting the shape of coronavirus.

Figure 1: (C) Cross-sectional view shows the major elements.

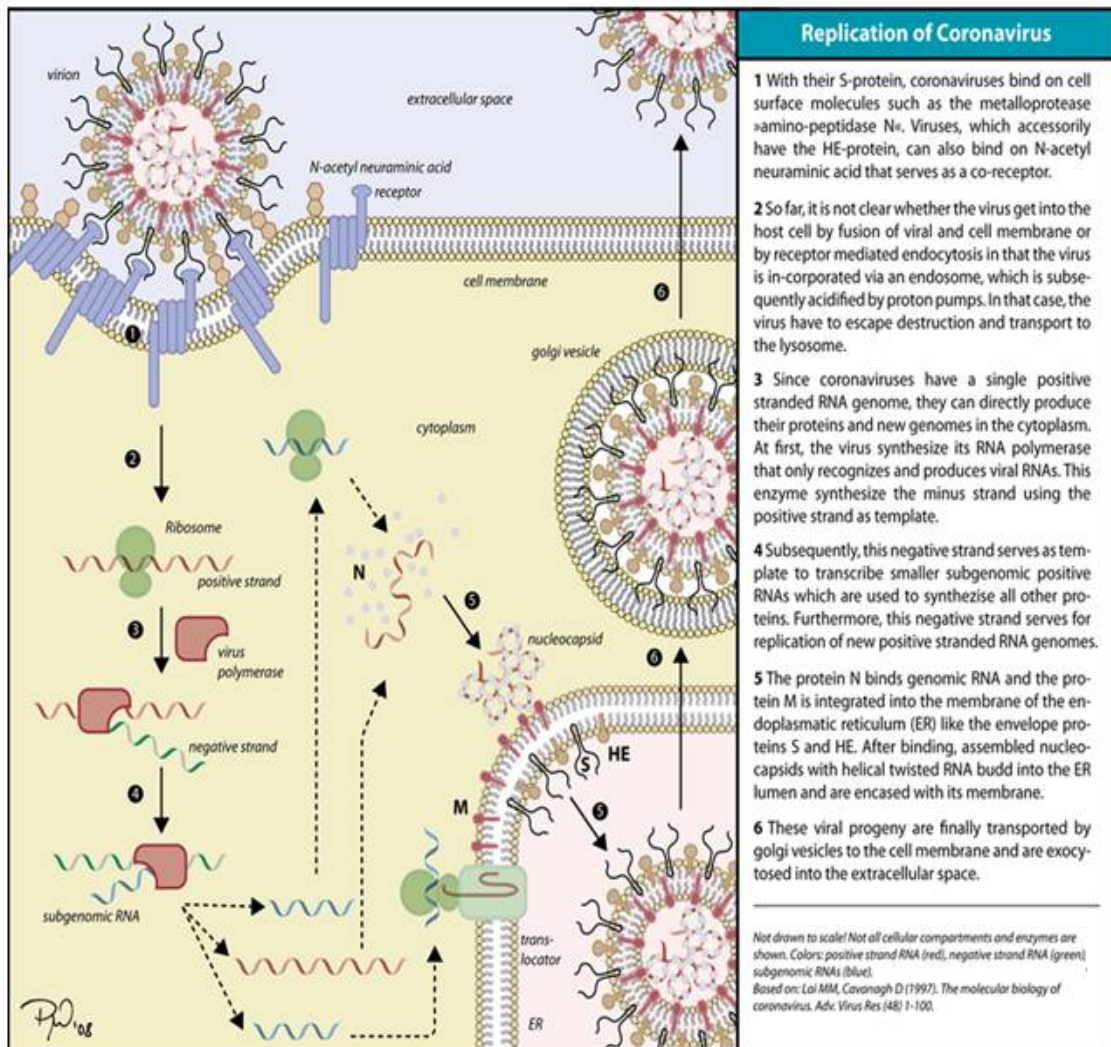


Figure 2: The infection cycle of a coronavirus¹³

Expression of Structure

Only the 5' most replicase gene is translated from the input positive-strand genome RNA. The genome contains multiple genes for the known structural proteins S, E, M, and N, as well as other genes for expression of proteins that have been labeled as "nonstructural" or "accessory" because they have been presumed not be required for replication. Moreover, they are thought not to be incorporated into virions. MHV encodes six of these genes, while SCoV encodes possibly up to 11 structural and accessory genes, which are expressed from subgenomic mRNAs¹⁴. Furthermore, the

deletion of a single extra character in individual isolates of SCoV, suggested that this character is concerned in the alteration for replication and transmission in humans¹⁵.

Genetics of Coronavirus

With the help of technological advances in the field of genetics, researchers have been able to learn about the pathogenesis and replication of CoV. The genetics of coronavirus replication and pathogenesis have largely been studied using natural variants, host range mutants, passaged virus, and mutagenized viruses selected for temperature sensitivity and specific phenotypes. Classical complementation of functions made it possible to define at least eight genetic groups for MHV, with most of the complementation groups localized to the replicase gene¹⁶. Research reveals that the major marker of tropism and pathogenesis is the S protein. Furthermore, the genetic segment in replicase may be responsible for the distinctive activities of coronavirus. Specific modification and groundbreaking range have been limited to studies of the 3' 10 kb of the MHV genome and restrict the selection of viable recombinants. The establishment of infectious clone reverse genetic strategies for the corona viruses transmissible gastroenteritis virus (TGEV); a better understanding of its genome has been made possible by utilizing "infectious clone" reverse genetic strategies¹⁷.

Gastroenteritis Coronavirus Transmissible

HCoV-229E, IBV, and MHV has made it theoretically possible to study the genetics of the entire genome and all of the structural, accessory, and replicase genes. Approaches to "infectious cloning" have included full-length cDNA clones of TGEV genome in bacterial artificial chromosomes. The in vitro assembly approach was developed to overcome the challenge of full-length cDNA cloning of the TGEV and MHV genomes, which contained "toxic" regions in the replicase gene, resulting in unstable or toxic clones in *E. coli*¹⁸. In combination with biochemical and cell imaging approaches, it is also possible to study highly defective or lethal mutations in electroporated cells in order to define critical determinants of replication. The in vitro assembly approach has been used to introduce marker mutations that are silent for replication in culture. Thus, direct reverse genetic studies of the critical replicase gene functions can be performed using in vitro assembly of infectious clones¹⁹.

Advances Research About SCoV

The initial research of coronaviruses that had been conducted by the National Institutes of Health, lead to the quick advancement in the recognition and study of SCoV as the etiologic instrument of SARS, in addition to the ability of the virus to viably grow in cultures²⁰. Corresponding knowledge of CoV was viable thanks to the swift detection of ACE II as the receptor for SARS; which verified the great importance of research of coronavirus subtypes. Likewise, replicase feature expression, handling and anticipated function have made it possible for therapeutic approaches and identification of genome structure studies²¹.

Reverse Genetics Studies Application

Robust tissue culture systems and emerging animal models, create the potential to rapidly answer questions concerning: (1) determinants of virus growth in culture; (2) potential mechanisms of transspecies adaptation; (3) sensitivity to and escape from biochemical and immune interference with replication; (4) determinants of virulence and pathogenesis; (5) mechanisms of genome recombination and mutation; (6) functions of and requirements for replicase, structural, and accessory proteins; and (7) development of stably attenuated viruses for use as seed stocks for inactivated vaccine or testing as live-attenuated vaccines²². While the Coronavirus genome and its protein makeup demonstrate great tolerance of modification, all sequences of quality-based insertion, deletion or mutagenesis, have so far answered to have viruses damage in replication, pathogenesis, or both. Large numbers of changes in MHV and other species of coronaviruses are observed in SCoV. This technique could be an attempt for likely attenuation in SCoV culture and animal models. Finally, it is essential to generate strains of SCoV that are weakened and stabilized against reversion and recombination; to discover factors for replication and pathogenesis determinants and construction of virus. Such weakened modifications would offer supplementary precautions in determining SCoV development, biology, disease, treatment, and prevention²³.

Coronaviruses in Human

Vaccines are components that are not therapeutic but protective. Seven strains of human coronaviruses have been identified (i) Human coronavirus HCoV-229E (ii) Human coronavirus HCoV-OC43 (iii) Severe acute respiratory syndrome

coronavirus SARS-CoV (iv) Human coronavirus HCoV-NL63, New Haven coronavirus (v) Human coronavirus HKU1 (vi) Middle East respiratory syndrome coronavirus MERS-CoV, previously known as novel coronavirus 2012 and HCoV-EMC. (vii) Severe acute respiratory disorder coronavirus SARS-CoV-2, previously known as nCoV-2019 or "novel coronavirus 2019". The coronaviruses HCoV-229E, - NL63, - OC43, and - HKU1 frequently move in the human population and cause respiratory infections in adults and children widely²⁴.

Table 1: Characteristics of patients who have been infected with SARS-CoV-2, MERS-CoV, and SARS-CoV²⁵

	SARS-CoV-2	MERS-CoV	SARS-CoV
Demographic			
Detection date	December 2019	June 2012	November 2002
Detection place	Wuhan, China	Jeddah, Saudi Arabia	Guangdong, China
Age average	49	56	39.9
Age range	21–76	14–94	1–91
Male:female ratio	2.7:1	3.3:1	1:1.25
Confirmed cases	80,423	2494	8096
Case fatality rate	2,708 (3.4%)	858 (37%)	744 (10%)
Health-care workers	16	9.8%	23.1%
Symptoms			
Fever	40 (98%)	98%	99–100%
Dry cough	31 (76%)	47%	29–75%
Dyspnoea	22 (55%)	72%	40–42%
Diarrhoea	1 (3%)	26%	20–25%
Sore throat	0	21%	13–25%
Ventilatory support	9.8%	80%	14–20%
Notes Data as of 21 January 2020.			






Conclusion

Coronavirus is highly pathogenic in nature. HCoVs causes serious intense respiratory disorder. For SARS-CoV and MERS-CoV, animal reservoirs were the source for the worldwide outbreak with shocking morbidity and mortality²⁶. The recognition of a new coronavirus identified in December 2019, named CoVID-19 are common for coronavirus researchers. CoVID-19 was documented in Wuhan, China, and has caused

severe infection and death globally. Some healthcare professionals have theorized that 2019-nCoV could progress to a low pathogenic yet highly infectious coronavirus, which may reoccur annually every winter such as the annual influenza virus. The critical expansion and outlook of this epidemic at the time of this writing is unknown as the virus is rapidly differentiating²⁷. Coronaviruses can be classified into four genus: alpha, beta, gamma, and delta, in which alpha and beta CoVs transmit the disease to humans. Four HCoV (HCoV 229E, OC43, NL63 and HKU1) are globally prevalent, all wide-ranging as evidenced by 30% of upper respiratory tract infections in adults²⁸. Coronaviruses are ecologically dissimilar with the most excellent diversity seen in bats, signifying that they are the main source in the production of large quantities of the virus. Warm-blooded animals are considered as an intermediate host; assisting in recombination and mutation extension of genetic variety. Normal symptoms of SARS, incorporate with pyrexia, coughing, dyspnea, and diarrhea. Thirty-two percent of infected patients require mechanical ventilation while 10% result in death²⁹. Increased fatality rates are observed in patients with medical comorbidities. Human to human transmission was evident chiefly in medical settings. The Nosocomial extension might be explained by basic virology. The major human receptor for the SARS S glycoprotein, the angiotensin-converting catalyst II (ACEII), is found chiefly in the lower respiratory tract, as opposed in the upper reparatory passage. MERS, SARS and the Wuhan coronavirus appear to cause more severe disease in older people, though uncertainty remains around the latest outbreak. Of the cases of Wuhan coronavirus reported so far, none are yet confirmed to be among children and old age group. Receptor sharing may represent upper respiratory tract signs and symptoms that top viral flaking occurring late into the tenth day of infection when the patient is hospitalized³⁰. SARS care often requires vaporized treatment protocols such as intubation, which may promote and increase nosocomial activity. The surface spike glycoprotein is dangerous for the adhering of host cell receptors and considered as a key determinant of host run restriction. HCoV has remained relatively low key due to their weak phenotypes in humans³¹. This changed in 2002, when cases of severe atypical pneumonia were described in Guangdong Province, China, causing worldwide concern as the disease spread via international travel to more than 202 countries. The disease got known as Severe Acute Respiratory Syndrome (SARS), and a beta HCoV, named SARS-CoV, was recognized as the causative factor. Since the new cases shared account of human-

animal contact at live game markets, zoonotic transmission of the virus was definitively supported. Palm civets and raccoon hounds were originally considered to be the animal reservoir. A few vital events occurred in select populations such as the well described mini flare-up in the Hotel Metropole in Hong Kong, where infected subjects traveled and spread SARS globally³². Another incident occurred at the Amoy Gardens housing complex where more than 310 people were infected, giving proof that air transmission of SARS-CoV may take place from time to time. About two decades later, the mechanism linked with the transmission of SARS-CoV, ranging from self-limited animal to human transmission to human–human transmission instance, remain unsuccessfully understood. Currently, while there is no treatment, researchers are at work. As is the case for most viral infections, symptoms usually subside independently. Health care providers suggest seeking professional care early. In the event that signs, and symptoms feel more severe than a standard cold, patients are recommended to seek immediate medical care. Physician can reduce side effects by prescribing pain or fever medication³³.

Table 2: A comparison of diagnostic criteria based on symptoms and travel³⁴

Symptoms	Coronavirus* (COVID-19) Symptoms range from mild to severe	Cold Gradual onset of symptoms	Flu Abrupt onset of symptoms	Seasonal Allergies Abrupt onset of symptoms
 Length of symptoms	7-25 days	Less than 14 days	7-14 days	Several weeks
 Cough	Common (usually dry)	Common (mild)	Common (usually dry)	Rare (usually dry unless it triggers asthma)
 Shortness of breath	Sometimes	No**	No**	No**
 Sneezing	No	Common	No	Common
 Runny or stuffy nose	Rare	Common	Sometimes	Common
 Sore throat	Sometimes	Common	Sometimes	Sometimes (usually mild)
 Fever	Common	Short fever period	Common	No
 Feeling tired	Sometimes	Sometimes	Common	Sometimes
 Headaches	Sometimes	Rare	Common	Sometimes (related to sinus pain)
 Body aches and pains	Sometimes	Common	Common	No
 Diarrhea	Rare	No	Sometimes for children	No

*Information is still evolving. **Allergies, colds and flus can all trigger asthma, which can lead to shortness of breath. COVID-19 is the only one associated with shortness of breath on its own. Sources: Asthma and Allergy Foundation of America, World Health Organization, Centers for Disease Control and Prevention

A room humidifier or a hot shower can help with a sore throat. Increased fluids consumption, rest and increasing the time of rest, all show to improve recovery³⁵. The Wuhan coronavirus mortality rate is greater than SARS and MERS, yet still similar to the 1918 Spanish influenza pandemic. Researchers believe the mortality rate is likely to be less because of an "iceberg" of milder cases are yet to be found. This highlights that novel viruses multiply far more rapidly through a population. Presently, a vaccine to safeguard against this family of viruses does not exist. Trials for a MERS vaccine are in progress. The US National Institutes of Health is working on a vaccine against the new species of virus. It will be a long time before clinical trials get in progress and greater than a year until they are made available. People may be able to reduce the risk of infection by avoiding people who are sick. Individuals must avoid touching the eyes, nose and mouth. Most people who become infected experience mild illness and recover, but it can be more severe for others. Washing the hands often with soap and water and for at least 20 seconds or cleansing with hand sanitizers may neutralize viruses that may be on the hands. Maintaining a social distance of at least 1 meter (3 feet) between yourself and anyone who is coughing or sneezing; as the mist of small liquid droplets from the nose or mouth may contain the virus. Because hands touch many surfaces and can pick up viruses; contaminated hands can transfer the virus through the mucous membrane of the eyes, nose or mouth and lead to pathogenesis in the host. The practice of positive respiratory hygiene includes covering your mouth and nose with a bent elbow or tissue when coughing and sneezing, and the immediate disposal of the used tissue. This practice should be encouraged and enforced in social communities. Following positive respiratory hygiene, hand washing/cleansing and avoidance of touching the eyes, nose and mouth, protect populations from viruses such as the cold, flu and COVID-19. Individuals with a fever, cough and dyspnea must seek early medical care. Individuals who feel ill should stay home. Individuals should follow advice given by healthcare providers and staying informed with the latest developments about COVID-19. Protective measures should be taken for individuals who are in or have recently visited (past 14 days) areas where COVID-19 is spreading³⁶. For diagnosis, most testing for COVID-19 is currently done on viral genetic material from nose and throat swabs using a workhorse tool of molecular biology known as reverse transcription-polymerase chain reaction (RT-PCR). Labs worldwide have customized their PCR tests for SARS-CoV-2, using different primers targeting different sections of

the virus's genetic sequence. Protective measures currently being applied by countries to combat the disease include the screening of travelers. The emergence of 2019-nCoV, has triggered health screenings at airports in multiple countries. The screenings will likely not be the deciding factor in whether the virus continues to spread globally. Quarantine means the separation of a person or group of people reasonably believed to have been exposed to a communicable disease but not yet symptomatic, from others who have not been exposed, in an effort to prevent the possible spread of the communicable disease. Currently, a fourteen-day quarantine is observed prior to days prior to an assessment in individuals suspected to have exposure. If traveling to China, learn the sign and symptom, maintain social distance of 3 meters and avoid settings such as live animal marketplace, which is where the latest breakout is thought to have originated in Wuhan. On December 31, 2019, Chinese authorities detailed groups of pneumonia cases in Wuhan, China where a large portion of the effected population was reported to have frequented live game markets. According to the current epidemiological situation in the world, the Coronavirus disease (COVID-19) outbreak has caused 509,164 human infections and 23,335 deaths spanning in 202 countries (WHO update: 27 March 2020). It is different from SARS-CoV in biological, epidemiological, and clinical highlights. Researchers at the Infectious Diseases Vaccine Research Center and the National Institute of Allergy, have made advances in the use of a nucleic acid vaccine as a possible treatment³⁷. During SARS, experts progressed in acquiring the genome of SARS-CoV to a stage I clinical trial of a DNA vaccine in 20 months and have since packed down that period to 3.5 months for other viral diseases. For 2019 nCoV, they desire to shift considerably faster, using representative RNA (mRNA) vaccine innovation³⁸. The international health authorities are now brainstorming on the search for therapies able to counter the most severe effects of the disease. In this regard 600mg of hydroxychloroquine have been found to be efficient on SARS-CoV-2 patients³⁹. In addition, it is also reported that Hydroxychloroquine is more effective with azithromycin and it causes significant reduction of the viral activity so it is concluded that combine therapy of Azithromycin with hydroxychloroquine significantly more efficient for virus elimination⁴⁰.

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