CHISOM MAYHELEN -ANAH SARS-CoV-2 RT-PCR POSITIVITY AMONG HEALTHCARE WORKERS. FEBRUARY, 2023

NEAR EAST UNIVERSITY

INSTITUTE OF GRADUATE STUDIES

DEPARTMENT OF MEDICAL MICROBIOLOGY AND CLINICAL MICROBIOLOGY

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MSc. THESIS

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I hereby declare that all information, documents, analysis and results in this thesis have been collected and presented according to the academic rules and ethical guidelines of Institute of Graduate Studies, Near East University. I also declare that as required by these rules and conduct, I have fully cited and referenced information and data that are not original to this study.

CHISOM MAYHELEN ANAH

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Thank you

CHISOM MAYHELEN ANAH

ABSTRACT

A STUDY ON SARS-COV-2 AND HEALTH CARE WORKERS IN NEAR EAST UNIVERSITY HOSPITAL, CHISOM MAYHELEN ANAH. INSTITUTE OF GRADUATE STUDIES, MEDICAL/CLINICAL MICROBIOLOGY PROGRAM, MASTER THESIS, NICOSIA, 2023.

This study aims to investigate the prevalence and risk factors of SARS-CoV-2 infection among healthcare workers (HCWs) at the Near East University Hospital in Nicosia, Cyprus. The study was conducted as a cross-sectional survey among the 763 HCWs working at the hospital, including physicians, nurses, and other healthcare professionals. Participants were asked about their demographics, medical history, and exposure to COVID-19 patients, followed by nasopharyngeal swab collection for SARS-CoV-2 detection by Real-Time PCR. The results showed that 197 of the HCWs tested positive for SARS-CoV-2, 26.9% were nurses and 14.2% were doctors. The study also identified several risk factors associated with SARS-CoV-2 infection among HCWs, including age, gender, smoking status, and comorbidities. The findings of this study highlight the importance of implementing effective infection control measures and providing adequate PPE for HCWs to prevent the transmission of SARS-CoV-2 in healthcare settings. Healthcare Workers and HCWs are used interchangeably.

KEYWORDS: sars-cov-2, hcws, source of infection, exposure.

ÖZ

CHISOM MAYHELEN ANAH. YAKIN DOĞU ÜNİVERSİTESİ HASTANESİ LİSANSÜSTÜ EĞİTİMLER ENSTİTÜSÜ TIP/KLİNİK MİKROBİYOLOJİ PROGRAMI, YÜKSEK LİSANS TEZİ, LEFKOŞA, 2023 SARS-COV-2 VE SAĞLIK ÇALIŞANLARI ÜZERİNE BİR ARAŞTIRMA.

Bu çalışma, Kıbrıs Lefkoşa'daki Yakın Doğu Üniversitesi Hastanesinde sağlık çalışanları (HCW'ler) arasında SARS-CoV-2 enfeksiyonunun prevalansını ve risk faktörlerini araştırmayı amaçlamaktadır. Çalışma, hastanede çalışan doktor, hemşire ve diğer sağlık profesyonellerini içeren 763 sağlık çalışanı arasında kesitsel bir anket olarak gerçekleştirilmiştir. Katılımcılara demografik bilgileri, tıbbi geçmişleri ve COVID-19 hastalarına maruz kalmaları soruldu, ardından RT-PCR ile SARS-CoV-2 tespiti için nazofaringeal sürüntü alındı. Sonuçlar, sağlık çalışanlarının 197'sinin SARS-CoV-2 için pozitif test yaptığını, %26,9'unun hemşire ve %14,2'sinin doktor olduğunu gösterdi. Çalışma ayrıca sağlık çalışanları arasında SARS-CoV-2 enfeksiyonu ile ilişkili yaş, cinsiyet, sigara içme durumu ve komorbiditeler dahil olmak üzere çeşitli risk faktörlerini de tanımladı. Bu çalışmanın bulguları, sağlık hizmeti ortamlarında SARS-CoV-2'nin bulaşmasını önlemek için etkili enfeksiyon kontrol önlemleri uygulamanın ve sağlık çalışanları için yeterli KKD sağlamanın önemini vurgulamaktadır. Sağlık Çalışanları ve Sağlık Çalışanları birbirinin yerine kullanılmaktadır.

ANAHTAR KELİMELER: sars-cov-2, sağlık çalışanları, enfeksiyon kaynağı, maruz kalma

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CHAPTER 1

INTRODUCTION

The word "coronavirus" derives from the Latin word "corona," which means "crown." The name derives from the virus's presentation under an electron microscope, where it looks as round particles completely covered by projections resembling the solar corona. Coronaviruses, which are positive-sense, encapsulated, single-stranded RNA viruses, were discovered in humans in 1965. Coronaviruses are a type of virus that has been linked to mild to moderate respiratory diseases among people. Three significant coronavirus outbreaks have since taken place: SARS-CoV in 2002, MERS-CoV in 2012, and the existing SARS-CoV-2 outbreak.(Cui et al., 2019)

On December 21st, 2019, the Chinese city of Wuhan revealed an emergence and spread of a pneumonia-like disease. On January 3, 2020, samples from sick people were taken and examined, uncovering a new coronavirus, orginally dubbed 2019-nCoVs by the Chinese Center for Disease Control and Prevention (CCDC) and later dubbed SARS-CoV-2 by the International Committee on Taxonomy of Viruses (ICTV).(Gorbalenya et al., 2019) This was in responding to the unexpected rise in cases and the confusion over medical care practices. The World Health Organization declared the outbreak a global epidemic on March 11, 2020, urging nations across the globe to take measures to safeguard their people.(WHO, 2020)

Due to their frontline status and lack of familiarity with the new coronavirus strain, healthcare workers (HCWs) have been recognized as one of the groups most at risk for exposure to COVID-19. The Chinese Centers for Disease Control and Prevention (CDC) reported in January 2020 that 16 healthcare workers (HCWs) had contracted COVID-19 from exposure to outbreak patients. Given the limited knowledge of the virus and the hazards that HCWs face when providing treatment, it is essential that they take extra precautions to prevent infection.(CDC, 2020)

It was suggested that infections among healthcare workers could potentially contribute to further transmission of the virus within hospitals and outside health facilities, making it crucial to prioritize protective protocols to safeguard them against COVID-19. The new coronavirus was given the name SARS-CoV-2 by the International Committee on Taxonomy of Viruses (ICTV) on February 11, 2020, while the disease was given the designation COVID-19 by the World Health Organization (WHO). Previously, this pathogen was referred to as 2019 new coronavirus (2019-nCov) or 2019 human coronavirus. (Human Coronavirus hCov-19 or HCV-19). The SARS pandemic of 2002–2004 was caused by SARS coronavirus type 2.(Gorbalenya et al., 2019)

Healthcare personnel are more vulnerable of getting infected compared to the general population, as the virus is extremely contagious and medical staffs are constantly exposed to the viral particles without adequate protective equipment.

Researchers around the world have sought to quantify the impact of COVID-19 on the general public, healthcare systems, and the challenges and dangers encountered by frontline healthcare providers. Additionally, a combination of stress, long working hours, and night shifts can make their immune systems more vulnerable.(Choi et al., 2022) For both SARS-CoV and SARS-CoV-2, the human angiotensin converting enzyme 2 (ACE-2) acts as the entry receptor, while human proteases are the entry activators. ACE-2 has been determined to be the functional receptor that facilitates SARS-CoV infection. Its nucleotide sequence is 79.0% identical to that of SARS-CoV and 51.8% identical to that of the Middle East Respiratory Syndrome Coronavirus (MERS-CoV). Both SARS-CoV and MERS-CoV were found in bats, and SARS-CoV-2 appears to be the same virus.(CDC, 2020) Due to their proximity to patients and lack of familiarity with this novel coronavirus strain, healthcare workers were soon recognized as one of the populations most at risk of contracting COVID-19.(Chou et al., 2020)

Currently, there are ongoing efforts in China to understand the origin of the COVID-19 outbreak. These investigations include studying early cases with symptoms that occurred near Wuhan in December 2019, sampling of the local environment and markets, and collecting information on the types of wildlife sold at the Huanan market and their potential role in the outbreak.(Worobey et al., 2022) It is known that coronaviruses primarily cause respiratory and gastrointestinal infections and are categorized into four types: Deltacoronavirus, Gammacoronavirus. Betacoronavirus, and

Alphacoronavirus.(Tao et al., 2021) Six types of human coronaviruses are known, including HCoVHKU1, HCoV-OC43, MERS-CoV, SARS-CoV, HCoV229E, and HCoV-NL63. While coronaviruses have been known to cause illness in humans, it was not until the 2002-2004 SARS outbreak, 2012 MERS outbreak, and the current COVID-19 pandemic that they gained widespread attention. Recent studies have suggested that bats are the original hosts of the SARS-CoV and MERS-CoV before being transmitted to other animals and eventually to humans.(Zhou et al., 2020)

Direct contact with an infected person, either by inhaled droplets or by touching an infected surface or object and subsequently contacting one's face, is the primary mode of transmission of COVID-19. Although it is feasible for COVID-19 to be spread through the air in some contexts, such as hospitals, where aerosol-generating procedures are being done, this is not currently thought to be a major method of transmission.(Q. Li et al., 2020) Evidence suggests the virus can be found in feces, however the significance of this finding in terms of transmission is unclear.

There have been 18,738,58 laboratory-confirmed cases of COVID-19 recorded around the world as of the second week of April 2020. People between the ages of 30 and 69 make up the bulk of the cases (77.8%). About three-quarters (77.0%) of the reported cases originated in China's Hubei province, where 21.6% are farmers or other workers.(Worobey et al., 2022)

CHAPTER 2

LITERATURE REVIEW

2.0 BACKGROUND

The COVID-19 pandemic is caused by the coronavirus subtype severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). The virus was initially termed "2019 novel coronavirus" and "human coronavirus 2019" in the beginning phases of the outbreak. The pathogen was deemed a Public Health Emergency of International Concern by the World Health Organization on January 30, 2020, and a pandemic by the end of 2020 following its discovery in Wuhan, China. SARS-CoV-2 has a positive-sense single-stranded RNA genome and is highly infectious.(Cw et al., 2019a)

It's a coronavirus belonging to the SARSr-CoV species and it's very similar to the virus that caused the SARS pandemic in 2002–2004. Due to its genetic similarity to bat coronaviruses, its likely etiological source is bats. It is still unclear if bats were the primary vector for SARS-CoV-2 transmission or if the virus was spread through other animals first. Due to the virus's lack of genetic diversity, it is believed that the initial outbreak that allowed human infection occurred in late 2019.(Y. Liu et al., 2020)

From December 2019 through September 2020, when no local residents were immune and no additional precautions were adopted, the number of new infections generated by each individual case of SARS-CoV-2, the virus that causes COVID-19, was expected to be between 2.4 and 3.4, according to epidemiological data.(Worobey et al., 2022) On the other side, it was found that some of the virus's variants were more contagious than the original. The virus is transmitted mostly through personal contact, but also by aerosols and respiratory droplets released through speech, breathing, and other forms of exhalation, as well as through coughing and sneezing. As a membrane protein that participates in the regulation of the renin-angiotensin system, it enters human cells through binding to the angiotensin-converting enzyme 2 (ACE2) receptor.(Q. Li et al., 2020)

2.1 TERMINOLOGY AND NAMES

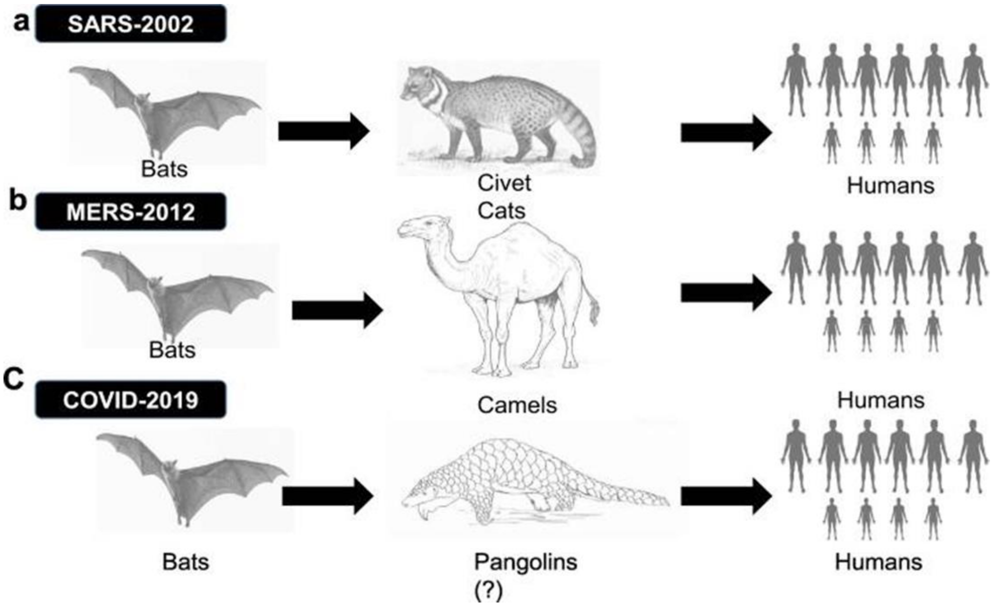
At first, the virus was referred to by a number of other names, including "the coronavirus" and "Wuhan coronavirus." In January 2020, however, the World Health Organization (WHO) suggested the interim term "2019 novel coronavirus" (2019-nCov) in order to comply with its 2015 guidelines prohibiting the use of specific locales, animal species, or human populations for naming diseases and viruses.(Q. Li et al., 2020)

During the earliest phases of the COVID-19 outbreak, the virus was referred to by a number of distinct names. These included "the coronavirus," "Wuhan coronavirus," and others. The World Health Organization (WHO) issued a recommendation for a provisional name for a newly identified coronavirus in January 2020; the name "2019 novel coronavirus" (2019-nCov) was chosen to comply with WHO recommendations that disease and virus names not reference specific locations, animals, or populations.(Worobey et al., 2022) This virus was officially designated as "severe acute respiratory syndrome coronavirus 2" by the International Committee on Taxonomy of Viruses on February 11, 2020. (SARS-CoV-2). In public health messaging, WHO often refers to SARS-CoV-2 as "the COVID-19 virus," and the designation HCoV-19 was inserted in certain scientific studies to distinguish it from the disease SARS. The World Health Organization has issued a statement calling "Wuhan virus" a harmful and xenophobic name for COVID-19.(Huang et al., 2020)

The Angiotensin Converting Enzyme-2 (ACE-2) protein is a human cell entry point for SARS-CoV and SARS-CoV-2, and both viruses require human proteases for replication. Evidence from multiple studies indicates that ACE-2 is the virus's functional receptor. Nucleotide sequence comparisons reveal that SARS-CoV-2 shares 79.0% similarity with SARS-CoV and 51.8% similarity with Middle East Respiratory Syndrome Coronavirus (MERS-CoV). SARS-CoV, as well as MERS-CoV, may have originated in bats, and there is speculation that SARS-CoV-2 originated in the same way.(Huang et al., 2020)

Figure 1.

THE ORIGINS OF CORONAVIRUSES



(Salahshoori et al., 2021)

2.2 TRANSMISSION AND INFECTION

In January 2020, it was revealed that SARS-CoV-2, the virus that causes COVID-19, has transmitted to humans, contributing to the current pandemic. At first, it was believed that the virus propagated predominantly through close-quarters respiratory droplet exchange, such as sneezing and coughing.(Q. Li et al., 2020) Although writing and typing are the most common modes of transmission, research has shown that speaking aloud and breathing infected aerosols could be equally effective. Between 200 and 800 viral particles are thought to be necessary for an infection to take hold.(C. Li et al., 2020) Concerns have been raised concerning the formation of aerosols from laboratory activities, which could put other people at danger, and this has important consequences for biosafety.

It is unclear how contagious the virus is while it is incubating; however, studies suggest that the pharynx, along with other parts of the upper respiratory tract, may contain the maximum viral load around day four post-infection or within the first week of symptoms. Furthermore, research suggests that SARS-CoV-2 RNA can remain in the body for as long as 46 days after the onset of symptoms. (CDC, 2020)

The nasal cavity may be the primary site of SARS-CoV-2 infection, according to research conducted at the University of North Carolina, with the virus moving to the lungs through aspiration. This research also discovered that the virus specifically targets ciliated cells and type 2 pneumocytes in the airways and alveolar regions of the lungs, and that there is a gradient of infection, with higher levels present in the proximal parts of the lungs and lower levels present in the distal parts.(Chua et al., 2020) Animals as diverse as cats, ferrets, hamsters, non-human primates, minks, tree shrews, raccoon dogs, fruit bats, and rabbits have all been shown to be susceptible to infection by SARS-CoV-2. That's why it's wise for those who've tested positive for the virus to keep their distance from animals/pets.(Chua et al., 2020)

2.3 ASYMPTOMATIC AND PRESYMPTOMATIC TRANSMISSION

The majority of people infected with SARS-CoV-2 may not show symptoms, and those people are 42% less likely to spread the virus. However, preparatory epidemiological simulation of the Chinese outbreak revealed that "pre-symptomatic shedding may be common among documented infections" and that subclinical infections may have caused the vast bulk of infections. A study of 94 hospitalized patients confirmed this finding, forecasting that people began shedding the virus 2-3 days before manifestations appeared. This implies that significant transmission may take place before there is an appearance of symptoms. Pertinently, more research is needed to fully comprehend the role of asymptomatic transmission in the propagation of SARS-CoV-2.(C. Li et al., 2020)

2.4 REINFECTION

It is unclear how often or for how long people develop immunity after a SARS-CoV-2 infection. Although the likelihood of reinfection is unknown, there have been examples of people being unwell to varied degrees after a second infection.

It is still unclear whether or not SARS-CoV-2 re-infection is possible. There have been reports, though, of people testing positive for the virus more than once. An early case in point is a 33-year-old Hong Kong man who tested positive on March 26th, 2020, then recovered and tested negative twice, and finally tested positive again on August 15th, 2020. Through whole-genome sequencing, researchers were able to determine that the viruses responsible for the two infections belonged to separate clades. Herd immunity may be difficult to acquire and vaccinations may not provide lasting protection if reinfection is possible, as suggested by these results.(To et al., 2020)

According to a separate study, a 25-year-old male from Nevada tested positive for SARS-CoV-2 on April 18th, 2020 and again on June 5th, 2020, despite having two negative tests in between. Genomic analysis confirming substantial genetic variations between the two virus strains established the case study as a reinfection. There is still a lack of understanding as to why this patient's second illness was so much worse than the first.(Tillett et al., 2021)

2.5 RESERVOIR AND ORIGIN

SARS-CoV-2, the virus responsible for COVID-19, has enigmatic origins. In 2019, Wuhan, China, saw the first confirmed cases. Yet, the virus's origin and its transition into human populations remain obscure. There was initial concern that the virus originated at the Huanan Seafood Market, where several of the first victims had worked.(Cui et al., 2019) But other studies point to tourists as the possible source of the virus's rapid spread in the marketplace. The World Health Organization (WHO) published a report in March 2021 stating that direct spillover from bats was the second most plausible reason after the virus crossing over to humans via an intermediate animal host. The Huanan Seafood Market and the food supply chain were ruled out as potential vectors for spread. (Zhou et al., 2020)

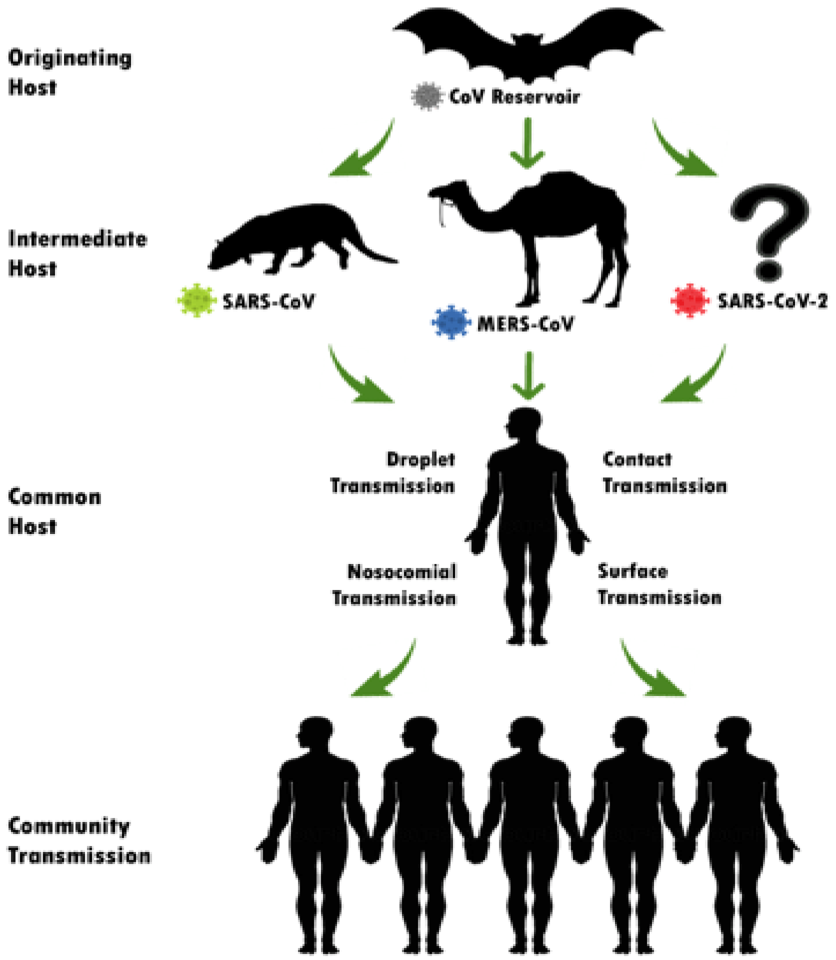
A study published in November 2021, however, revealed that the first known case had been misinterpreted and that the majority of early cases related to the Huanan Market supported it as the source. Studies on the development of SARS-CoV-2, the virus responsible for COVID-19, are underway.(Worobey et al., 2022) It stands to reason that a virus that has only recently been acquired by interspecies transmission will show signs of fast development. Researchers have calculated that SARS-CoV-2 undergoes a mutation rate of roughly 6.54 x 10-4 per site per year. SARS-replication CoV-2's machinery is thought to limit the virus's ability to evolve rapidly despite coronaviruses' notoriously strong genetic flexibility.(Amicone et al., 2022)

Multiple coronaviruses with a high degree of similarity to SARS-CoV-2 have been identified in bat populations as part of the investigation into the outbreak's source. These coronaviruses were collected from several species of bats in Laos and were found there. Another study indicated that of the viruses studied, the one discovered in Chinese bats (RaTG13) was the most similar to SARS-CoV-2 (96.1% similarity). It is important to note, however, that none of the aforementioned viruses is believed to be SARS-direct CoV-2's ancestor.(Amicone et al., 2022)

Although SARS-CoV-2 (the causative agent of COVID-19) has a murky past, bats are generally accepted as the most plausible natural reservoir of the virus. It is possible, nevertheless, that the virus was passed on to humans from an intermediary host. There was speculation that pangolins might have contributed to the spillover occurrence, although this has not been confirmed by further study. Studies have showed that pangolin virus samples have a weak binding affinity for the human ACE2 receptor and have only a 92% sequence identity with SARS-CoV-2. No one knows for sure how the virus got into human beings.(X. Li et al., 2020)

Figure 2.

CORONAVIRUS GENESIS AND TRANSMISSION CONCEPT



(Sheam et al., 2020)

2.6 CLASSIFICATION

SARS-CoV-2 is a beta (β) genus member of the subfamily Coronavirinae and family Coronaviridae, which is a group of viruses that are largely animal in origin. The alpha and beta coronaviruses' natural host is the bat, and more specifically the fruit bat, while the gamma and delta coronaviruses' natural host is the pig or the bird.(Fan et al., 2019) The SARS-CoV-2 is the seventh human-infecting coronavirus, joining 229E, NL63, OC43, HKU1, MERS-CoV, and the original SARS-CoV. From the common cold to deadly infections like Middle East respiratory syndrome (MERS), these viruses are responsible for a wide spectrum of disorders.(Adachi et al., 2020)

COVID-19, caused by SARS-CoV-2, is caused by a coronavirus and is classified in the β-coronavirus genus. Whole genome sequencing led to this categorization. Bats, and more specifically fruit bats, are thought to be the natural hosts of coronaviruses belonging to the alpha and beta subtypes, whereas pigs and birds are the natural hosts of coronaviruses belonging to the gamma and delta subtypes. Remember that this data is still being studied and may change in the future. Analysis of the complete genome of SARS-CoV-2 confirmed that it is a member of the β-coronavirus family and, more specifically, the β -coronavirus genus.(Ar Gouilh et al., 2018) It has a single, linear RNA segment and is a positive-sense, single-stranded RNA virus. Both mammals and birds are susceptible to infection by coronaviruses, not just humans. In humans, coronaviruses can produce a wide range of symptoms, from the ordinary cold to the extremely dangerous and lethal Middle East respiratory disease (MERS). After 229E, NL63, OC43, HKU1, MERS-CoV, and the original SARS-CoV, SARS-CoV-2 is the eighth human-infecting coronavirus. (Lu et al., 2020)

Similar to SARS-CoV (the virus responsible for the 2003 SARS outbreak) and MERS-CoV (the virus responsible for the Middle East respiratory syndrome), SARS-CoV-2 (the virus responsible for COVID-19) is a member of the coronavirus family. Sarbecovirus B is a beta-coronavirus, and it belongs to the subgenus Sarbecovirus. Most coronaviruses, including SARS-CoV-2, are very adaptable genetically and can frequently recombine via copy-choice replication.(Gorbalenya et al., 2019) The RNA sequence of this virus is around 30,000 bases long, which is quite a bit for a coronavirus. Similar to other coronaviruses, its genome is dominated by regions that code for proteins.(Lu et al., 2020)

The polybasic site that is cleaved by the furin protease is a defining feature of SARS-CoV-2. The pathogenicity of the virus is thought to be enhanced by this property. Researchers believe that SARS-capacity CoV-2's to infect humans is related to the virus's acquisition of a furin-cleavage site in its S protein.(Lu et al., 2020) In SARS-CoV-2, the amino acid sequence upstream of arginine and serine, which compose the

S1/S2 cleavage site of the spike protein, is the furin protease's recognition site. Similar sites are widespread among members of the Orthocoronavirinae subfamily of viruses, but they are unusual in the Beta-CoV genus and are found nowhere else in the SARS-CoV-2 subgenus. SARS-CoV-2, like the feline coronavirus, is an alpha-coronavirus, and they share a furin cleavage site.(Gorbalenya et al., 2019)

Phylogenetic research, which includes comparing the viral sequences of various genomes, can be used to learn more about SARS-genetic CoV-2's makeup. Experts can piece together the virus's evolutionary history by studying several different genomes. Early in the year 2020, some virus genomes had already been extracted and examined by groups like the Chinese Center for Disease Control. By the month's conclusion, we had significantly increased the number of genomes we had studied to 42.(Gorbalenya et al., 2019)

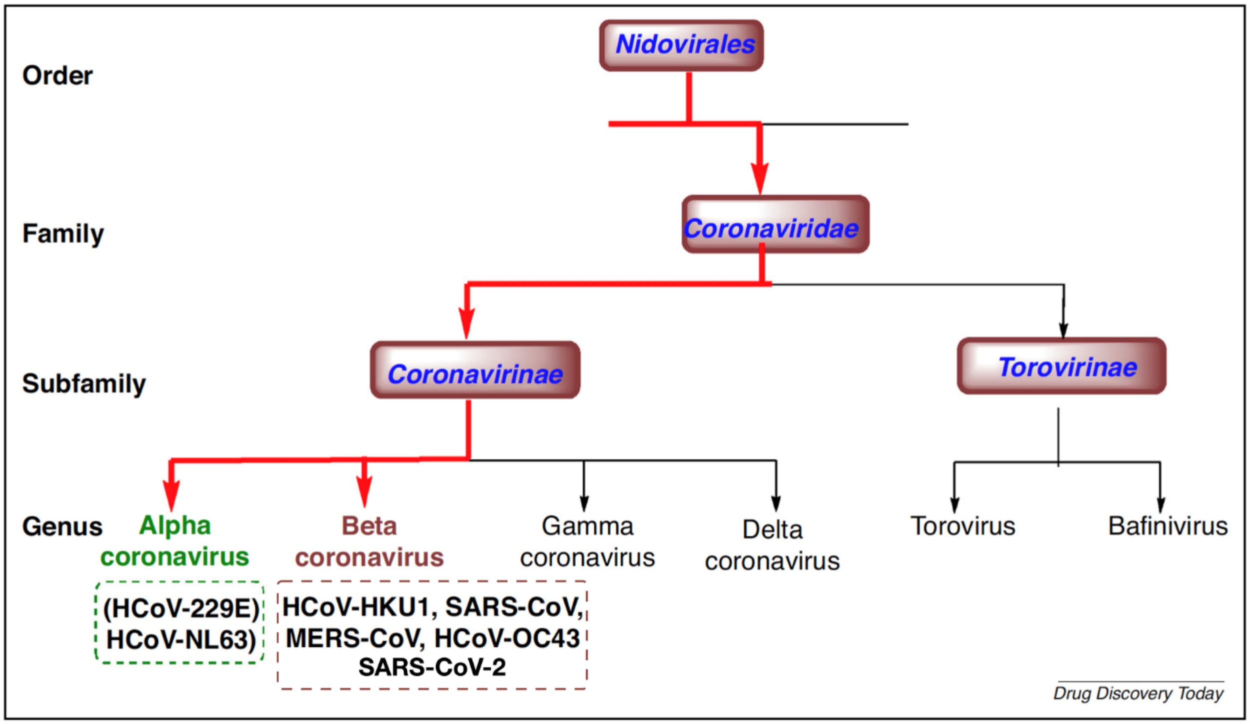
The genetic makeup of SARS-CoV-2 can shed light on important questions about the virus's genesis and dissemination. With enough sequenced genomes and phylogenetic analysis, scientists may build a tree depicting the virus's mutation history. The Chinese Center for Disease Control and other universities had evaluated five SARS-CoV-2 genomes as of January 12, 2020.(A. Wu et al., 2020a) There were no more than seven alterations between these samples and their common parent, suggesting that the first human infection happened in late 2019. As of August 2021, the genomes of 3,422 SARS-CoV-2 strains were published online. These strains were collected from every continent except Antarctica. In a report published on 11 February 2020, the International Committee on Taxonomy of Viruses determined that 2019-nCoV, the virus responsible for COVID-19, is a member of the coronavirus family related to severe acute respiratory syndrome (SARS). In addition, researchers stated in July 2020 that a highly contagious form of SARS-CoV-2, designated G614, had emerged as the primary strain during the pandemic. This was determined by comparing the viruses' DNA sequences using a rule set that examines the associations between coronaviruses based on five conserved nucleic acid sequences.(Cui et al., 2019)

Open reading frames (ORFs) are regions of DNA that can be translated into proteins, and coronaviruses like SARS-CoV-2 have six of these ORFs in their genetic makeup.

A putative overlapping gene, designated ORF3d, was found in the SARS-CoV-2 genome in October of 2020. Although its precise role has yet to be established, it has been shown to provoke a robust immunological reaction. Previous research has linked this gene to a coronavirus strain that causes disease in pangolins. However, greater investigation into ORF3d's function in SARS-CoV-2 is required.(A. Wu et al., 2020a)

Figure 3.

CLASSIFICATION OF CORONAVIRUS



(Zhu et al., 2020)

2.7 VARIANTS

It's vital to keep in mind that multiple SARS-CoV-2 variants have emerged, and these variants can be organized into bigger groups called clades. Despite the fact that these variants are classified in a variety of ways by various organizations, a few stand out. The World Health Organization (WHO) has identified five variations as having "urgent" health consequences and is closely monitoring their development.(He et al., 2023) Remember that virology is a constantly developing discipline, therefore any and all data presented here may soon be outdated. Currently, there are five varieties that the WHO considers to be cause for alarm.

- The UK-discovered Alpha variety (B.1.1.7), which includes changes in its spike protein that could make it more contagious and resistant to immunity from previous infection or vaccination.
- The South African-discovered beta variation (B.1.351) has changes in its spike protein that may increase its virulence and resistance to immunity from previous infection or vaccination.
- The gamma variation (P.1) was discovered first in Brazil; it features several mutations in the spike protein that may increase its transmissibility and resistance to immunity from previous infection or vaccination.
- Delta variant (B.1.617.2), which was initially discovered in India and contains several mutations in the spike protein that may make it more transmissible and resistant to immunity from past infection or vaccination.
- Some changes in the spike protein of the Japanese-discovered Kappa variant (B.1.1.28) may increase its virulence and resistance to immunity conferred by previous infection or vaccination.

The aforementioned variants' properties are currently being studied; thus, they may change as new information is gathered.(Tao et al., 2021)

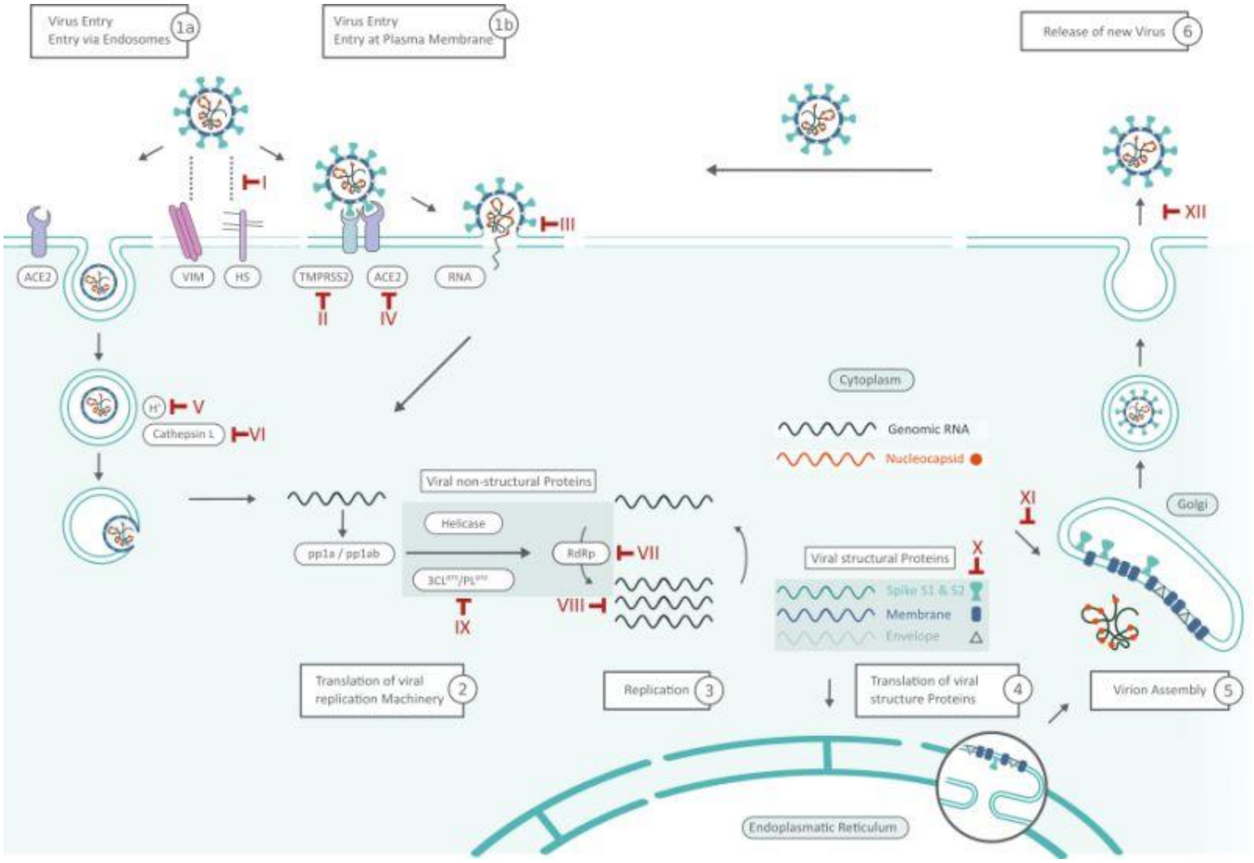
2.8 VIROLOGY

The virus responsible for SARS-CoV-2, also known as COVID-19, is a single-stranded RNA virus. Protein-coding sequences make up the vast majority of the virus's genome, which is around 30,000 base pairs in length. The virulence, host range, and immune response of a virus are all determined by its genetic makeup. Attachment to the host cell occurs when the spike protein of SARS-CoV-2 binds to the ACE2 receptor on the surface of the host cell, kicking off the replication cycle.(Salahshoori et al., 2021) When a virus infects a cell, it takes use of the cell's replication mechanism to make copies of itself. The spike protein, envelope protein, membrane protein, and nucleocapsid protein are all components of SARS-CoV-2. These proteins contribute to viral replication and host cell adhesion, and they also form the virus's physical shape.

Since the spike protein is responsible for binding to the ACE2 receptor on host cells, it plays a crucial role in facilitating viral entry and infection.(Salahshoori et al., 2021)

Figure 4.

SARS-COV-2 LIFE CYCLE



(Hoffmann et al., 2020)

2.9 GENOME

There are four main open reading frames (ORFs) in the SARS-CoV-2 genome, and they all code for different sets of proteins. ORF1ab, the first ORF, encodes 16 non-structural proteins required for viral replication. These proteins aid the virus in both cloning its genetic material and resisting the host immune system.(A. Wu et al., 2020b)

It has been proven that the protein encoded by the second ORF, ORF3a, contributes to viral proliferation and pathogenicity. The ORF3 is known as ORF6, and it encodes a protein that is involved in viral replication and host cell entrance.(A. Wu et al., 2020b)

Four structural proteins, including the spike (S) protein, envelope (E) protein, membrane (M) protein, and nucleocapsid (N) protein, are encoded by the fourth ORF, which is also known as ORF7a-ORF10. Among these proteins, the spike protein is the most well-known since it is the protein responsible for binding to the ACE2 receptor on the surface of the host cell and thereby allowing the virus to enter the host cell.(A. Wu et al., 2020a)

The leader sequence, the trailer sequence, and the poly-A tail are all parts of the SARS-CoV-2 genome that play critical roles in the virus's replication and spread. It is believed that the trailer sequence aids in packaging the virus, and the leader sequence is involved in viral reproduction. There is speculation that the poly-A tail controls the stability of viral RNA.(Lu et al., 2020)

As a whole, the SARS-CoV-2 genome is a complicated and multifunctional entity that is essential for viral replication, host cell entrance, and immune evasion. Accurate treatment and vaccination against COVID-19 require a thorough understanding of the virus's DNA.(A. Wu et al., 2020a)

2.10 REPLICATION CYCLE

The first step in SARS-CoV-2 replication is the virus' attachment to the ACE2 receptor on the surface of the host cell. The viral genome is released into the host cell's cytoplasm after the virus has successfully infected the host cell. Host ribosomes use the viral DNA to produce viral proteins. New infectious particles, or virion, are assembled from the viral proteins and discharged from the host cell.(Y. Liu et al., 2020)

It is possible to categorize the steps involved in the virus's replication cycle into four broad categories: attachment, entry, replication, and release. When the virus connects to the ACE2 receptor on the surface of the host cell, this is known as the attachment phase. When a virus invades a host cell, this is known as the entry phase. The replication phase involves the translation of the viral genome into viral proteins and the assembly of new virions. Once the new virions have been produced, they must be released from the host cell, which is the release step.(Yang et al., 2019)

The infectious potential of a virus depends on many factors, including the virus's genetic makeup, replication cycle, and structural proteins. Due to its genetic make-up, the virus is able to avoid detection by the host's immune system and attach itself to the ACE2 receptor. The virus can infect new hosts thanks to its ability to replicate.(Majchrzak & Poręba, 2022)

As reiterated earlier, COVID-19 is caused by the SARS-CoV-2 virus, which enters host cells via the ACE2 receptor and replicates there. Once inside the cytoplasm of the host cell, the virus's genetic material, a single-stranded positive-sense RNA, can begin to replicate. The ribosomes of the host cell use the viral RNA as a template to synthesize a polyprotein, which is then cleaved by proteases produced by the virus.(Bajrai et al., 2022)

One of the first proteins to be expressed is the replicase-transcriptase complex, which uses the viral RNA as a template to produce more viral RNA, a process called transcription. The newly synthesized viral RNA is then used to synthesize more viral proteins, including the structural proteins that make up the viral envelope.(Majchrzak & Poręba, 2022)

The viral envelope, or spike, protein binds to the ACE2 receptor on host cells and allows the virus to enter. Once inside, the virus's replication cycle continues. The virus's replicase-transcriptase complex continues to produce more viral RNA, which is then used to produce more viral proteins. The newly synthesized viral proteins, including the spike protein, are assembled into new virus particles.(Bajrai et al., 2022)

The virus then buds out of the host cell and is released into the surrounding environment, where it can infect other cells. The virus's replication cycle is highly efficient, allowing it to produce millions of new virus particles in a single infected cell. The efficiency of the virus's replication cycle is one of the reasons why COVID-19 is so contagious.(Y. Liu et al., 2020)

It is worth noting that the virus also has a mechanism to protect its genetic material from host's defense mechanism, known as the viral RNA-dependent RNA polymerase (RdRp), which plays a central role in the replication of the viral genome by synthesizing a full-length, positive-strand RNA from the viral genomic RNA template. The viral proteases cleave the polyprotein into individual functional proteins, including the viral RdRp.(Yang et al., 2019)

2.11 VIRAL PROPERTIES AND CHARACTERISTICS

The coronavirus family, of which SARS-CoV-2 is a part, also includes other respiratory viruses like the common cold. The RNA of the virus is linear and has a little over 30,000 bases; it is a positive-sense, single-stranded RNA (+ssRNA) virus. 11 proteins are encoded by the virus's 29,903-nucleotide genome, including the spike (S), envelope (E), membrane (M), nucleocapsid (N), and open reading frames 1a, 1b, 2, 3a, 3b, 3c, and 3d.(Majchrzak & Poręba, 2022)

Because it interacts to the ACE2 receptor on human cells and facilitates viral entry, the spike protein is the virus's most crucial protein. The nucleocapsid protein attaches to the viral RNA to create a compact structure, while the envelope and membrane proteins provide a protective outer coating. Other proteins perform tasks like replication, viral life cycle regulation, and host immune response.(K. Liu et al., 2020)

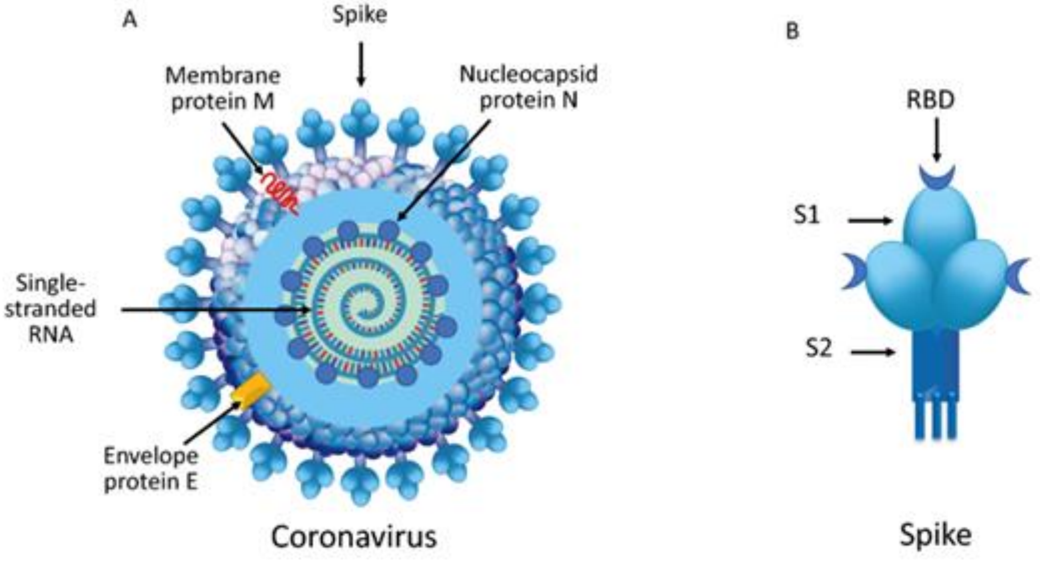
The virus copies its genetic material and synthesizes viral proteins in the cytoplasm of infected cells by hijacking the host cell's machinery. Attachment of the spike protein to the ACE2 receptor on the host cell initiates the replication cycle, which is followed by the release of the viral genetic material into the host cell.(Lu et al., 2020) Afterward, the viral RNA is utilized as a blueprint to produce new copies of the virus's protein coat and genetic material. Following their synthesis, the freshly constructed viral particles are discharged from the host cell.(Q. Li et al., 2020)

It's possible for the virus to undergo mutation and evolution, resulting in the formation of new strains. Five variations have been identified by the WHO as being of particular concern due to their potential to increase transmission and/or resistance to existing vaccinations. These variations are called B.1.1.7, B.1.351, P.1, P.2, and L452R. To sum up, SARS-CoV-2 is a highly contagious virus that can result in life-threatening respiratory infection. It is important to consider the genetic make-up and replication cycle of the virus.(Peiris et al., 2003)

The image below shows the (a) structural proteins membrane (M), envelope (E), and spike (S) of coronavirus anchored to the viral envelope, which contains the ribonucleoprotein core, i.e., the nucleocapsid protein (N), which acts as a scaffold around the single-stranded RNA. (b) The surface spike is made up of the S1 subunit, which contains the receptor binding domain (RBD), and the S2 subunit, which serves as the stem, anchoring the spike to the viral envelope and allowing host cell fusion after protease activation.

Figure 5.

STRUCTURE OF CORONAVIRUS



(Rossi et al., 2020)

2.12 PATHOPHYSIOLOGY

SARS-CoV-2 (the virus that causes COVID-19) pathogenesis is poorly known. However, researchers have uncovered some of the virus's most crucial pathways for causing illness in humans. The ability of COVID-19 to infect and reproduce in the upper respiratory tract of humans is a major element in the pathophysiology of the virus. Viruses infect humans by entering the respiratory system and binding to cell surface receptors there. Once within, the virus replicates to create more infectious virus particles. Coughing, fever, and breathlessness are only some of the symptoms caused by the virus as it replicates and irritates and damages the cells lining the respiratory system.(Adachi et al., 2020)

COVID-19's ability to trigger an internal "cytokine storm" is also crucial to understanding the pathophysiology of the virus. When the immune system creates an abundance of inflammatory proteins called cytokines, this is known as a cytokine storm. These cytokines are normally produced in response to infection and help to fight the virus. However, in some cases, the immune system produces too many cytokines, leading to a severe and sometimes deadly overreaction. This overreaction can cause widespread inflammation, damage to vital organs, and even organ failure.(Cui et al., 2019; Rossi et al., 2020)

In addition to these mechanisms, the virus also causes changes in the body's blood vessels, leading to blood clots and other problems that can cause severe complications such as stroke, heart attack, and organ failure. This can happen in many ways, such as by direct infection of the endothelial cells lining the blood vessels, by activating clotting mechanisms or by overwhelming the immune system leading to clotting in small vessels.(Rossi et al., 2020)

Additional difficulties may arise as a result of the virus's ability to infect and multiply in other organs, including the brain, heart, and kidneys. The virus can cause serious illness and death in some people while having no effect at all, or just moderate symptoms, in others. Age, coexisting disorders, and genetics all have a role in determining how severely a disease manifests in a given individual.(Yu & Malik Peiris, 2005)

The pathophysiology of COVID-19 is, in short, complex and poorly understood. Scientists have only recently begun to understand how the virus causes disease in humans, but they have already pinpointed several key mechanisms, such as the virus's ability to infect and replicate in the respiratory tract, its ability to cause a "cytokine storm" in the body, its ability to cause changes in the body's blood vessels, and its ability to infect and replicate in other organs. The age of the patient, the presence of other medical disorders, and the individual's genetic makeup all have a role in the disease's progression and severity.(Cui et al., 2019)

2.13 CLINICAL CHARACTERISTICS

COVID-19 might cause mild to severe symptoms. Fever, coughing, shortness of breath, exhaustion, body aches, and a loss of taste and smell are among the most often reported symptoms. Even if they don't have any symptoms themselves, some people can still infect others and transmit the virus. COVID-19 has an incubation period of 2-14 days (but as little as a few hours and as much as 24 days).(Tao et al., 2021) Most people start feeling sick within 5 or 6 days after being exposed. Pneumonia, ARDS, septic shock, and multiple organ failure are all possible outcomes of a severe case of COVID-19. In addition to triggering stroke, heart attack, and deep vein thrombosis, the virus can also induce blood clots (DVT).(Huang et al., 2020)

People with certain preexisting disorders, such diabetes, heart disease, obesity, or HIV/AIDS, as well as the elderly and those with compromised immune systems are at a far higher risk of developing life-threatening illnesses. Droplets expelled by a coughing or sneezing infected person are the most common vector for the virus's propagation. Touching an infected surface or object and then touching your eyes, nose, or mouth is another way to transfer the virus.(Hoffmann et al., 2020)

Multiple tests, including as polymerase chain reaction (PCR), antigen testing, and serologic testing, can be used to diagnose COVID-19. PCR testing is commonly used for diagnosis confirmation because it is widely regarded as the most accurate procedure. Components of supportive care for COVID-19 include oxygen therapy and hydration control. Medications like remdesivir, which work by inhibiting the production of new viruses, have been demonstrated to reduce the length of time certain people are ill. Dexamethasone and other steroid medications have been shown to significantly lower mortality rates in seriously ill individuals.(K. Liu et al., 2020)

Preventative vaccines against COVID-19 have been produced and are available for use in an emergency. These vaccines protect against infection by priming the immune system to recognize and destroy the virus. Evidence shows that the immunizations are quite successful at preventing serious disease and death from COVID-19.(Tao et al., 2021) Overall, SARS-CoV-2 is a highly contagious virus that can manifest itself in a broad variety of ways. It can cause serious disease and even death in certain people. Droplets exhaled through the nose and throat are the most common vector for the virus's propagation, and it can be detected using a number of different procedures. To prevent infection, vaccinations are available, and supportive care is the mainstay of treatment.(Tao et al., 2021)

2.14 EPIDEMIOLOGY

SARS-CoV-2 was first detected in humans in December 2019 in Wuhan, China. By January 2020, the virus had already gone beyond China's borders, with instances being reported in Thailand, Japan, South Korea, and the United States. Pandemic status was announced by the World Health Organization on March 11, 2020, and the outbreak was labeled a Public Health Emergency of International Concern on January 30, 2020.(Zhu et al., 2020)

It was hypothesized that bats served as the natural reservoir for SARS-CoV-2 and that this was the mode of transmission through which the virus was first spread. The virus was then believed to have been transferred to humans by an intermediary host, such as a pangolin. However, further research has been unable to verify pangolins supposed intermediate host status.(Fan et al., 2019)

SARS-CoV-2 is spread from person to person through respiratory droplets spread by coughing, sneezing, and talking. Contact with infected surfaces or objects, as well as breathing air in a building with poor ventilation, are additional vectors for the virus's propagation.(Yu & Malik Peiris, 2005)

The incubation period of SARS-CoV-2 is typically 2-14 days, with the majority of persons displaying symptoms within 5 days after infection. Although the virus is most contagious when the patient is showing symptoms, it can also be spread by others who aren't showing any signs of illness.(Rowe et al., 2004)

There are a variety of factors that have affected the spread of SARS-CoV-2, such as population density, travel habits, and immunity levels. Cities and nursing homes, both of which have high populations, have been hit particularly hard by the virus's quick spread. The global spread of the virus has also been aided by international travel.(Peiris et al., 2003)

Older persons, vital workers, and people of color have all been hit worse by the virus than the general population. These differences have their roots in factors including preexisting conditions, occupational choices, and socioeconomic standing. The results suggest that some minorities have been disproportionately affected by the COVID-19 epidemic. Several variables, such as socioeconomic and structural inequality and health disparities, contribute to this.(Rowe et al., 2004)

Socioeconomic inequality is a significant contributor to the disproportionate impact of COVID-19 on some racial and ethnic groups. Some minority groups, especially the Latino and Black communities, may be more susceptible to the virus since they are more likely to live in poverty and have less access to healthcare. It's important to note that minorities are disproportionately represented in high-risk occupations like those in health care and retail trade, where they are more likely to be exposed to the virus.(Salahshoori et al., 2021)

COVID-19 disproportionately affects some racial and ethnic groups because of underlying systemic inequalities. This includes discrimination and racism in housing, education, and employment, which can lead to overcrowded living conditions, inadequate access to healthcare, and limited job opportunities, all of which can increase the risk of infection and poor outcomes from the virus.(Salahshoori et al., 2021)

In response to the pandemic, public health interventions have been implemented at the local, national, and international levels. These interventions include measures such as physical distancing, mask-wearing, testing, contact tracing, quarantine, and vaccination. The epidemiology of SARS-CoV-2 continues to evolve as the virus spreads and new information becomes available. Ongoing surveillance and research are critical for understanding the current and future impact of the virus on public health, and for guiding effective response efforts.(Lu et al., 2020)

2.15 ROUTES OF TRANSMISSION

SARS-CoV-2, the virus responsible for COVID-19, is spread mostly by the airborne droplets produced by an infected person while coughing, sneezing, talking, or breathing. Any person in close proximity can catch an infection from these droplets if they inhale them or come into touch with their eyes, nose, or mouth. The virus can also be passed from person to person through touching an infected surface or object and then touching one's own face.(Q. Li et al., 2020)

Close contact with an infected individual, such as in a hug or a handshake, can also transfer the infection. Close contact transmission describes this phenomenon. The airborne virus can easily spread in confined settings with inadequate air circulation. Contacting an infected surface or object and then touching one's own mouth, nose, or eyes is another way to transfer the virus. Fomite transmission describes this phenomenon. Depending on the surface and environmental conditions, the virus can remain viable for several hours or even days.(Y. Liu et al., 2020)

The airborne transmission of the virus is another mode of transmission, particularly in poorly ventilated indoor environments. Airborne transmission is the technical term for this phenomenon. In poorly ventilated locations or areas with a high number of sick persons, the virus can remain in the air for several minutes or even hours. The danger of transmission is highest in closed places, where people are in close proximity, and when people are not using masks or other protective equipment. Long-term care institutions and congregate living environments, where people live closely together, also increase the chance of transmission.(Yang et al., 2019)

Contact with infected surfaces or items can potentially spread the virus, a process known as fomite transmission. This is a less prevalent mode of transmission than respiratory droplet transmission, but it is nevertheless a mode of dissemination. Viruses can also be dispersed through the air. Small droplets called aerosols can be discharged into the air whenever an infected person speaks, coughs, or sneezes, and can stay there for minutes to hours, especially in poorly ventilated areas.(Rothe et al., 2020)

Infection can spread in two ways: through inhalation of airborne microbes, or through contact with fomites (infected surfaces or objects). The virus can survive on many different surfaces, according to a study published in the New England Journal of Medicine. This raises the prospect of indirect transmission via fomites and aerosolization. Following COVID-19 patients, it was observed that causes other than respiratory droplet dispersion may have contributed to some occurrences of transmission.

Despite quarantine efforts, it was found viral RNA on a variety of surfaces in the cabins of both symptomatic and asymptomatic persons, suggesting a possible role for fomites in transmission. Detection of ambient viral shedding on surfaces in a quarantine situation is consistent with the likelihood of indirect contamination and airborne transmission, notably, there is a lack of information on the virus's transmissibility because no live virus has been recovered from fomite samples.(Q. Li et al., 2020)

RESPIRATORY DROPLETS: The SARS-CoV-2 virus is spread mostly by respiratory droplets. Speaking, coughing, and sneezing all result in the release of these droplets from an infected person. They can be breathed either by those in close proximity to them or by those who come into contact with them after touching surfaces that the particles have settled on. The droplets have a diameter of more than 5 microns and can travel up to or more than 1 meter in the air. There is a greater chance of inhalation because they might linger in the air for a while. Aerosolization reduces the size of respiratory droplets so that they can be suspended in the air for longer, allowing them to be inhaled by humans at greater distances.(Q. Li et al., 2020)

CLOSE CONTACT: SARS-CoV-2 can also be spread by extremely close contact, which is defined as being within 2 meters of an infected person for at least 15 minutes. Although this is a much less typical route of transmission than through respiratory droplets, it can still happen if an infected person is in close quarters with others and is talking, coughing, or sneezing.(Rothe et al., 2020)

FOMITE: The SARS coronavirus type 2 (SARS-CoV-2) can be spread from person to person via fomite transmission, in which the virus is passed from an infected surface or object to a human host. One way this can happen is if an infected individual touches a surface and then someone else touches their face after touching the same surface. Fomites can take many forms, including everyday items like door knobs, elevator buttons, shopping carts, and mobile phones.(Q. Li et al., 2020)

AIRBORNE: The World Health Organization (WHO) and many other health organizations have said that SARS-CoV-2 is primarily transmitted by respiratory droplets, not through the air, which raises questions about the validity of claims that the virus can be spread through breathing. Some scientists, however, have hypothesized that the virus could be spread by airborne particles that would remain in the atmosphere for extended periods of time. This can happen in crowded indoor places with limited ventilation, such as hospitals, care facilities, and overcrowded dwellings.(Q. Li et al., 2020)

FECAL-ORAL ROUTE: When the virus is present in the feces of an infected person and that person’s feces are afterwards consumed by a healthy person, this is known as the fecal-oral route. This can happen if an infected individual touches food or other objects that another person eats without first thoroughly washing their hands after using the restroom.(Q. Li et al., 2020)

2.16 VERTICAL ROUTE OF TRANSMISSION

The transmission of SARS-CoV-2 from an infected mother to her child through the placenta, the placenta accreta, or breast milk is known as the vertical route of transmission. Evidence suggests the virus can be passed vertically through a mother's blood, urine, amniotic fluid, and breast milk. More study is need to properly grasp the scope of the vertical transmission risk, however, the probability of vertical transmission during pregnancy is modest, with only 2% of babies born to mothers infected with COVID-19, according to a study by the Centers for Disease Control and Prevention (CDC). Since there have been so few known cases of infants getting the virus during delivery, the danger of vertical transmission is likewise regarded to be low.(Q. Li et al., 2020)

Since the virus is rarely discovered in breast milk, breastfeeding is not thought to be a major mode of transmission. More studies are needed to completely understand the dangers and potential outcomes for infants born to mothers with COVID-19 because of the lack of data on vertical transmission. Expectant mothers should take precautions against contracting the virus by acquiring the COVID-19 vaccine and practicing good hygiene during the labor and delivery process.(He et al., 2023)

In 2020, Li et al. did another investigation into the vertical spread of SARS-CoV-2. The purpose of this study was to examine the incidence of SARS-CoV-2 in pregnant women and their infants. Of the 1,297 pregnant women who were tested for SARS-CoV-2, 20 (1.54%) tested positive. There were 14 vaginal births and 6 c-sections among the group of 20 women. Within 12 hours of birth, all of the babies were tested for SARS-CoV-2, and all of them came back negative. Further research is needed to corroborate the study's finding, but the danger of vertical transmission from mother to newborn appears to be low. (Li, et al., 2020)

The potential effects of COVID-19 pregnancies on newborns are mainly unknown. In a study of 27 Chinese pregnant women infected with SARS-CoV-2, 2 of the babies tested positive, suggesting vertical transmission from mother to child. The study's modest sample size, however, means that more investigation into the effects of SARS-CoV-2 on newborns and the possibility of vertical transmission is warranted. Vertical transmission of SARS-CoV-2 was found to occur in just 2 (1.0%) of 191 pregnant women in another investigation with a larger sample size conducted in Wuhan, China. Only 1.2% of the newborns had questionable outcomes, and the vast majority (96.8%) were born free of SARS-CoV-2 infection. These investigations demonstrate the feasibility but low frequency of vertical transmission.(Zhu et al., 2020)

2.17 SEXUAL ROUTE OF TRANSMISSION

The risk of sexual transmission of SARS-CoV-2, the virus that causes COVID-19, is minimal in comparison to other routes of transmission, such as respiratory droplets or close contact, yet sexual transmission has been recorded in a small number of cases.(D. Li et al., 2020)

It is possible to spread SARS-CoV-2 by any kind of sexual contact, including the genital area, the ano-rectal area, and the oral cavity. Sex between a guy who had recovered from COVID-19 and a woman caused her to contract the virus. The virus was confirmed, and the woman experienced minimal symptoms. Another example involved a female carrier who transmitted the COVID-19 virus to her male spouse through her vaginal secretions. The man was infected and had only minimal symptoms.(Song et al., 2020)

Sexual transmission of SARS-CoV-2 is thought to be low but remains unclear. The virus has been found in vaginal secretions and sperm, but how long it may live in these environments and whether or not it can cause illness is unknown. The virus has been shown to live in semen for up to two weeks in some investigations, and for up to three months in others. Individuals who have recently recovered from COVID-19 may still be at increased risk of sexually transmitting SARS-CoV-2 because to viral shedding in the semen or vaginal secretions. There is an increased chance of transmission due to the presence of the virus in pre-ejaculate or pre-vaginal secretions.(Song et al., 2020)

Safer sex practices, such as using condoms or dental dams, avoiding intercourse with partners who have symptoms of COVID-19, and limiting the number of sexual partners, are recommended by experts to lower the risk of sexual transmission of SARS-CoV-2. Since the risk of sexual transmission may be higher for those who have recently recovered with COVID-19, they may wish to delay engaging in sexual activity until they are sure they are no longer infected.(D. Li et al., 2020)

It's also crucial to remember that despite widespread belief that SARS-CoV-2 cannot be spread sexually, it is still possible for the virus to be passed on in this way. As a result, people need to be alert to the danger and take measures to safeguard themselves and their partners. Because the virus could affect developing babies and lead to serious disease, pregnant women should exercise extra caution. Notably, 9 of the patients who tested positive for SARS-CoV-2 also tested positive in their semen samples suggesting a high likelihood of sexual route of transmission.(D. Li et al., 2020)

In 2020, Li et al. conducted research into the possibility of sexual transmission of SARS-CoV-2 among humans. Researchers enlisted 101 couples, each with one certified COVID-19 positive and one negative member. All couples were told to avoid physical contact and to use condoms during sexual activity because the positive partners were experiencing mild to moderate symptoms. After 14 days of observation, the study discovered that among the 101 couples, one of the initially negative partners was actually positive for SARS-CoV-2. Samples of sperm from 14 of the positive partners were obtained and examined, with the results showing that viral RNA was present in the sperm of 5 of the 14 males (or 35.7%). More research is needed to confirm this and to understand the precise processes of sexual transmission, but the study does imply that there may be a potential for sexual transmission of SARS-CoV-2.(D. Li et al., 2020)

The goal of a 2020 cross-sectional investigation published in JAMA Network Open was to determine how often SARS-CoV-2 RNA was in the vaginal and spermatic fluids of people with COVID-19. In total, 87 people took part in the trial, and 42 of them had detectable levels of SARS-CoV-2. Researchers identified SARS-CoV-2 RNA in the sperm of 3 (7.4%) of the 42 people who tested positive for the virus, but in none of the 45 people who tested negative. Furthermore, no RNA from SARS-CoV-2 was found in vaginal secretions.(Khusid et al., 2020)

Virus shedding in semen was not linked to infectiousness, and the study authors concluded that the prevalence of SARS-CoV-2 in semen was low. They also noted the importance of further study to determine the likelihood of SARS-CoV-2 sexual transmission and the consequences for public health policies.(D. Li et al., 2020)

It should be noted that the sample size of this study was rather limited, thus more research is needed to confirm the findings and to better understand the risk of sexual transmission of SARS-CoV-2 and its possible consequences for public health guidelines.(D. Li et al., 2020)

2.18 OCULAR ROUTE OF TRANSMISSION

The transmission of SARS-CoV-2 through the eyes has not been thoroughly researched as of yet. Conjunctivitis, keratoconjunctivitis, and uveitis are just few of the ocular signs that have been observed in COVID-19 individuals. This indicates that the virus may infect the ocular surface, either directly through respiratory droplets or indirectly through contact with contaminated hands or objects.(Sun et al., 2020)

Conjunctivitis was the initial symptom of SARS-CoV-2 infection, according to research in the Journal of Medical Virology in 2020. Viral RNA was detected in both the conjunctival and throat swabs, indicating that the patient had contracted the virus through the ocular surface. The authors of the study also noticed that the patient showed no signs of respiratory illness, lending further credence to the idea that the disease was spread through the patient's eyes.(P. Wu et al., 2020)

In 2020, researchers reported four examples of COVID-19 patients who exhibited ocular signs in a study published in the British Journal of Ophthalmology. Three out of four individuals with conjunctivitis or keratoconjunctivitis had the virus found in ocular swabs. The researchers hypothesized that the virus probably entered the eye via respiratory droplets or infected hands or objects.(Sun et al., 2020)

Conjunctivitis and keratoconjunctivitis were among the ocular symptoms experienced by 11 of the 19 individuals with COVID who were studied in a case series published in the New England Journal of Medicine in 2020. The researchers found that all of the patients' ocular swabs contained viral RNA, and that in certain cases, the virus was also present in the patients' respiratory swabs. Transmission to the eyes was hypothesized to have occurred by respiratory droplets or via contact with contaminated hands or objects.(P. Wu et al., 2020)

Although there is currently just a small amount of study on the possibility of SARS-CoV-2 infecting the ocular surface, these investigations raise the possibility that this may be possible. More study is required to determine the likelihood of ocular transmission and to create effective preventative and therapeutic measures for people with COVID-19 who exhibit ocular signs.(Sun et al., 2020)

However, these studies only use a small sample size, thus more investigation into the possibility of ocular transmission is warranted. Those who have been exposed to or diagnosed with COVID-19 should take steps to prevent the spread of the virus through respiratory droplets, contact with contaminated surfaces, or contaminated hands. Good hand hygiene, covering one's face, and not touching one's face are all part of this.(Sun et al., 2020)

The transmission of SARS-CoV-2 through the eye has not been thoroughly researched as of yet. However, COVID-19 patients have been documented to experience a variety of ocular symptoms, such as conjunctivitis, keratoconjunctivitis, and uveitis. This indicates that the virus may infect the ocular surface, either directly through respiratory droplets or indirectly through contact with contaminated hands or objects.(Sun et al., 2020)

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Conjunctivitis and keratoconjunctivitis were among the ocular symptoms experienced by 11 of the 19 individuals with COVID who were studied in a case series published in the New England Journal of Medicine in 2020. Ocular swabs from all patients contained viral RNA, and in certain cases, the virus was also detected in respiratory swabs, as reported by the study's authors. Transmission to the eyes was hypothesized to have occurred by respiratory droplets or via contact with contaminated hands or objects.(Mungmungpuntipantip & Wiwanitkit, 2020)

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However, these studies only use a small sample size, thus more investigation into the possibility of ocular transmission is warranted. Individuals with COVID-19 symptoms or confirmed cases should take steps to prevent the spread of the virus by respiratory droplets, contact with contaminated surfaces, or contaminated hands. Good hand hygiene, covering one's face, and not touching one's face are all part of this.

2.19 SARS COV-2 AND HEALTHCARE WORKERS

Due to their significant patient contact, healthcare workers (HCWs) have a greater risk of catching SARS-CoV-2 than the general public. The overall seroprevalence of SARS-CoV-2 among HCWs was 15.4%, with a range of 0-42.9% across different studies and regions, according to a systematic evaluation of 2020 studies. The seroprevalence among healthcare workers was highest in intensive care units (ICUs; 31.2%), next in emergency rooms (EDs; 22.5%), and finally in inpatient wards (17.1%).(Yamada et al., 2021)

The absence of PPE is a major risk factor for SARS-CoV-2 infection among HCWs (PPE). Researchers in the United States discovered that HCWs who said they did not always use PPE were more likely to test positive for SARS-CoV-2 than those who said they did. Health care workers (HCWs) who reported not always wearing PPE (such as a mask or gown) were shown to have a greater likelihood of testing positive for SARS-CoV-2 than HCWs who did.(Therese et al., 2020)

The absence of adequate infection control procedures is another risk factor for the spread of SARS-CoV-2 among HCWs. Based on the results of an Indian investigation, healthcare workers who said they did not regularly practice hand hygiene were more likely to test positive for SARS-CoV-2 than those who said they did. Another American study indicated that healthcare workers who said they did not regularly exercise respiratory hygiene were more likely to test positive for SARS-CoV-2 than those who said they did so regularly.(Vicentini et al., 2021)

Health care workers infected with SARS-CoV-2 may face an increased risk of mortality and serious disease. HCWs who tested positive for SARS-CoV-2 in a U.S research(Gershon et al., 1995) had a higher risk of hospitalization and mortality than the general population. Medical personnel who tested positive for SARS-CoV-2 in another Italian study were more likely to be hospitalized to the intensive care unit (ICU) and to die than the general population.(Cw et al., 2019a)

Essential infection control measures, such as the correct use of PPE and adherence to hand hygiene and respiratory hygiene standards, are needed to protect HCWs from SARS-CoV-2 infection. Healthcare workers (HCWs) should have access to diagnostic tests, quarantine and isolation protocols, and counseling services.(Gershon et al., 1995)

Since the beginning of the COVID-19 pandemic, there has been substantial investigation and concern over the epidemiology of SARS-CoV-2 infection among healthcare workers (HCWs). Due to their high patient volume, HCWs are at higher risk of getting SARS-CoV-2 due to exposure to the virus in the form of respiratory droplets, close contact, and contact with contaminated surfaces or equipment.(Cw et al., 2019a)

High prevalence of SARS-CoV-2 among HCWs has been observed by several investigations. Physicians and nurses had the greatest rates of infection among HCWs, according to a study conducted in Wuhan, China.(Sugimoto & Kohama, 2020) In a comparable study in Italy, researchers discovered that 15% of healthcare workers were infected with SARS-CoV-2; the risk of infection was highest in those who worked in intensive care and emergency rooms.(Dowlatshahi et al., 2020)

Infection rates among healthcare workers have been found to be lower in another research. An American investigation indicated that whereas most cases of SARS-CoV-2 were asymptomatic, only 3% of HCWs tested positive for the virus.(Chou et al., 2020) Among healthcare workers in Spain, only 2% tested positive for SARS-CoV-2, and those who did were mostly asymptomatic or had minor symptoms, according to research published in 2012.(Borras-Bermejo et al., 2020)

HCWs in high-risk settings, such as intensive care units, emergency departments, and others where there is a high possibility of interaction with infected patients, appear to be at the greatest risk of contracting SARS-CoV-2. There is an elevated risk of SARS-CoV-2 infection among HCWs who work in high-volume settings, with patients who have tested positive for SARS-CoV-2, or who have had prolonged, close contact with infected patients or coworkers.(Chou et al., 2020)

Healthcare workers (HCWs) face a higher risk of contracting SARS-CoV-2 for a number of reasons. One theory proposes that this is because HCWs are more likely to come into touch with infected patients and be exposed to contaminated surfaces and equipment in the course of their work. Another theory suggests that the stress and long hours endured by HCWs may make them more susceptible to SARS-CoV-2 infection and more severely ill.(Dowlatshahi et al., 2020)

In conclusion, HCWs were disproportionately affected by the SARS-CoV-2 pandemic, bearing a higher risk of infection and severe disease than the overall population. This emphasizes the need for efficient infection control methods and the provision of sufficient resources for HCWs to safeguard their health and well-being.(Dowlatshahi et al., 2020)

2.20 PSYCHOLOGICAL IMPACTS OF SARS-COV-2 ON HEALTH CARE WORKERS

The SARS-CoV-2 pandemic has had a profound effect on the psyches of healthcare professionals. Stress, burnout, and mental health problems have worsened since the pandemic began, among other difficulties.

Stress is one of the most noticeable psychological effects of SARS-CoV-2 on healthcare professionals. Professionals in the medical field have been on the front lines of the pandemic, putting in extra hours and coping with a massive influx of patients. Stress and exhaustion have increased as a result, which might have harmful impacts on their wellbeing. When compared to the general population, healthcare workers report significantly greater levels of stress, burnout, and depression, according to a study done by the World Health Organization (WHO).(Cw et al., 2019a)

SARS-CoV-2 also has the potential to have an emotional toll by increasing the rate of burnout among healthcare providers. Medical staff have become emotionally drained due to the lengthy and severe nature of the pandemic. Symptoms of burnout include hopelessness, cynicism, and disengagement from one's work. Nurses have reported high levels of burnout and emotional tiredness as a result of the epidemic, according to research undertaken by the International Council of Nurses (ICN).(Cw et al., 2019b)

SARS-CoV-2 has important psychological effects on healthcare providers due, in part, to the possibility of mental health problems. Anxiety and sadness are among the mental health problems that can be exacerbated by the widespread uncertainty and dread caused by the pandemic. Healthcare workers have reported greater rates of anxiety and depression compared to the general population, according to research undertaken by the National Center for Biotechnology Information (NCBI).(Borras-Bermejo et al., 2020)

The mental health of medical staff has also been affected by the quarantine and isolation measures taken to prevent the spread of the virus. Having to be apart from friends and family has made me feel lonely and alone. Healthcare workers have reported increased feelings of loneliness and isolation as a result of the epidemic, according to a study published in the Journal of the American Medical Association (JAMA).(Shanafelt et al., 2020)

Numerous studies have detailed the emotional toll that SARS-CoV-2 takes on those who work in healthcare. Stress, burnout, and mental health problems have all increased significantly since the pandemic began.

Researchers in the United States discovered that medical professionals had an increased prevalence of psychological health conditions such as depression, anxiety, and post-traumatic stress disorder (PTSD) than the entire populace. According to the study, direct care providers for COVID-19 patients were more likely to feel psychological distress than those who were not engaged in care delivery.(Rajkumar & Sampathkumar, 2020)

According to another Chinese study, healthcare workers there had a greater rate of mental health issues like depression, anxiety, and insomnia than the general population. Frontline healthcare personnel were more likely to experience psychological discomfort than those who were not directly involved in patient care, according to the study.(Lai et al., 2020)

In addition to the emotional toll the epidemic has taken on healthcare personnel, it has also led to an alarming rise in burnout. In the United States, researchers discovered that individuals who worked directly with COVID-19 patients were more likely to experience burnout than those who did not.

Concerns have been made concerning the potential psychological impact of SARS-CoV-2 on healthcare workers. To help their employees cope with the emotional toll of the epidemic, healthcare companies should offer counseling and other services. Included in this category are programs that help people support and learn from one another as well as educational materials for stress reduction and overall mental health.(Lai et al., 2020)

In conclusion, the SARS-CoV-2 pandemic has had a significant impact on the mental well-being of medical professionals. Researchers have found that medical professionals are much more susceptible than the overall population to experience psychological health problems such as depression, anxiety, and post-traumatic stress disorder. They are also more likely to experience burnout. To help their employees cope with the emotional toll of the epidemic, healthcare companies should offer counseling and other services.(Shanafelt et al., 2020)

2.21 VIOLENCE AGAINST HEALTHCARE WORKERS DURING COVID-19

There is burgeoning worry that the COVID-19 outbreak will result in an increase in violence against healthcare workers. There has been an upsurge in violence towards healthcare personnel as a result of the stress, worry, and anxiety brought on by the pandemic and the difficulties of caring for patients with a highly dangerous disease. There are a wide variety of violent behaviors that fall under this umbrella.

Several countries have reported cases of physical violence against healthcare professionals due to the pandemic. It has been reported that patients and their family have punched, kicked, and spat at healthcare professionals in the United States. The effects of this kind of assault on healthcare workers' health and safety can be devastating.(Choi et al., 2022) Healthcare staff have also reported experiencing verbal abuse and harassment during the pandemic. Verbal abuse can take many forms, including threats and name-calling. Medical staff exposed to such violence are at a higher risk of acquiring psychological issues such as worry and depression. There is a complicated web of causes behind the uptick in violence towards healthcare professionals that has been observed during the pandemic. Personal protection equipment (PPE) for healthcare professionals is in short supply and has also been noted as a contributing factor to the spread of the pandemic's stress and panic among healthcare personnel.(Lai et al., 2020)

The government and healthcare sectors have responded with a variety of initiatives aimed at addressing this problem. Provide personal protection equipment and other safeguards, as well as mental health services, for those who work in the healthcare industry. Unfortunately, despite these measures, violence towards healthcare workers has been a major issue during the current COVID-19 outbreak. Governments and healthcare organizations must maintain collaboration to find solutions to this problem and ensure the safety of those who work in the healthcare sector.(Choi et al., 2022)

CHAPTER 3

3. AIM

The goals of this study were to (1) understand comprehensively the positivity of SARS-CoV-2 among healthcare workers, including the factors that raise the risk of infection to reduce this risk (2) spot potential sources of infection both inside and outside of the professional setting.

3.1 **MATERIALS AND METHODS**

3.1.1 STUDY GROUP

Northern Cyprus' Near East University Hospital served as the site for this retrospective investigation. Participants included were healthcare professionals who were tested for COVID-19 by polymerase chain reaction between March 2020 and February 2022. Information about the subjects were recorded, such as their ages, sexes, countries of origin, ailments they suffer from.

3.1.2 TOOLS AND EQUIPMENTS

- SARS-CoV-2 Real-Time PCR:

  A regular PCR kit that double gene RT-qPCRs (DIAGNOVITAL (RTA Laboratories Inc, DIAGNOVITAL SARS-CoV-2 PCR Kit, Istanbul- Turkey) and BioSpeedy, Bioeksen Inc, Istanbul- Turkey) will be utilized in SARS-CoV-2 diagnosis.
- Personal Protective Equipment (PPE):

  Equipment used by healthcare workers to protect themselves and patients against infection, such as gloves, gowns, masks, and face shields.
- Swabs:

  For COVID-19 testing, nasopharyngeal, oropharyngeal, and mid-turbinate swabs are utilized to collect upper respiratory tract samples.
- Specimen collection tubes:

  These tubes are used to transfer and preserve COVID-19 testing samples acquired from patients.

- Centrifuges:

  These devices separate various components of a material, such as DNA or RNA, for use in diagnostic tests.
- Nucleic acid extraction kits:

  They are used for extracting RNA or DNA from patient samples to be utilized in diagnostic procedures such as polymerase chain reaction (PCR).
- PCR machines:

  They are used to amplify and identify the presence of certain DNA or RNA sequences, such as SARS-CoV-2.

3.1.3 METHODOLOGY

This presents a comprehensive description of the methods involved in SARS-CoV-2 real-time PCR, a widely utilized approach for COVID-19 detection. The thesis will go over sample collection, RNA extraction and purification techniques, reverse transcription, real-time PCR setup, cycle parameters, and methods for data analysis in detail. This thesis will aid in the comprehension of the benefits and limits of SARS-CoV-2 real-time PCR for COVID-19 diagnosis by analyzing each of these phases thoroughly in Chapter 5.

- Collection of samples: A nasopharyngeal swab is used to acquire a respiratory sample from the patient.
- RNA extraction: Using a commercial nucleic acid extraction kit, the viral RNA is extracted from the sample. After that, the extracted RNA is purified and concentrated.
- Reverse transcription: With reverse transcriptase, the extracted RNA is transformed to complementary DNA (cDNA).
- Real-time PCR setup: The cDNA is combined with PCR reaction components such as specialized primers and probes that target SARS-CoV-2 genomic regions. The mixture is then poured into a PCR plate and placed in a real-time PCR machine.

- Real-time PCR cycling: The PCR machine cycles through many temperature changes, allowing the target SARS-CoV-2 DNA sequence to be amplified. The fluorescent signal released by the probe attached to the target DNA sequence is measured throughout each cycle.
- Data analysis: The real-time PCR machine generates data that may be evaluated to determine the presence of the target SARS-CoV-2 DNA sequence in the sample. The cycle threshold (Ct) value is used to determine how much viral RNA is present in a sample. The test result is deemed positive for SARS-CoV-2 if the Ct value is less than a specified threshold. The test result is negative if the Ct value is greater than the threshold. Overall, real-time PCR is a very highly sensitive and specific method for detecting SARS-CoV-2, and it is currently one of the most extensively used methods for diagnosing COVID-19.

Figure 6.

DIAGNOVITAL® SARS-CoV-2 Real Time PCR Kit

3.2 DATA COLLECTION AND ANALYSIS

This study enlisted 197 individuals, and their demographic information was recorded. The demographic information of the participants was acquired and kept on an Excel spreadsheet, including their names, barcodes, identification/passport numbers, gender, nationality, date of birth, age, phone numbers, and addresses.

The statistical analysis used was SPSS (Statistical Package of the Social Sciences) Demo Ver 22 (SPSS Inc., Chicago, IL, USA). A program used for all statistical analysis of the data, in order to determine statistical significance, Pearson Chi-square, Fisher's Exact Test, and One-Way ANOVA tests were used and $p<0.05$ values were considered as significant. Ethics committee approval of the study was obtained with the project number NEU/2022/100-1501 at the meeting held by the NEU Scientific Research Ethics Committee on 24/02/2022.

In this study, the inclusion criteria are:

- Participants must be healthcare professionals affiliated with the Near East University Hospital in Northern Cyprus.
- Participants must have tested positive for COVID-19 using polymerase chain reaction (PCR) between July 2020 and December 2022. and also, HCWs that tested negative so as to make statistical comparison.

CHAPTER 4

4.1 RESULTS

The study population comprised 763 healthcare workers employed at a university hospital between 1 July 2020 and 31 December 2022, and SARS CoV-2 PCR positivity was assessed between these dates.

There were 259 (33.9%) men & 504 (66.1%) women in the study cohort. Throughout the study period, the rate of COVID-19 infection among healthcare professionals was 25.8% (197/763).

The first table illustrates the occupational distribution of the persons in the study group in the hospital. Individual professions and COVID-19 infection were shown to have no connection (p=0.435). Also, there was no significant association between gender and COVID-19 (p=0.844).

Table 1. Distribution Of Participants Gender and Age

GENDER	**NUMBER**	**%**	**STANDARD DEVIATION**	**MEAN**
MALE	68	34.5	11.705	36.49
FEMALE	129	65.5	10.177	35.78
TOTAL	197	100	10.704	36.03

The distribution of the male and female gender involved in this study is detailed in **TABLE.1** using descriptive statistics. The mean age of the healthcare workers with positive PCR testing was 36.03±10.70 with 68 (34.5%) being male and 129 (65.5%) being female (between 21-69 years). When the gender and mean age of the positive patients were compared, the variation between the averages was found to be insignificant (p=0.663).

Table 2. The Occupations of The Healthcare Workers in The Hospital

POSITION	**NUMBER**	**PERCENTAGE (%)**
DOCTOR	104	23.9
NURSE	182	13.6
OTHERS	477	62.5
TOTAL	763	100

TABLE.2 above shows data containing the total account of HCWs that work in Near East Hospital. The other occupations that were not listed in the table above include; Patient counselor, cleaning staff, carer, porter, paramedic, administrative, laboratory - radiology staff, operating room technicians etc.

Table 3. Contingency Table of PCR Positivity and Occupation

		OCCUPATION			
		NURSE/CARETAKER	**DOCTOR**	**OTHER**	**TOTAL**
PCR - Positive	**Count**	**53**	**28**	**116**	**197**
	Expected Count	**47.0**	**26.9**	**123.2**	**197.0**
	Among PCR patients%	**26.9%**	**14.2%**	**58.9%**	**100.0%**
	Among same occupation%	**29.1%**	**26.9%**	**24.3%**	**25.8%**
Negative	**Count**	**129**	**76**	**361**	**566**
	Expected Count	**135.0**	**77.1**	**353.8**	**566.0**
	Among negative PCR patients%	**22.8%**	**13.4%**	**63.8%**	**100.0%**
	Among same occupation%	**70.9%**	**73.1%**	**75.7%**	**74.2%**
Total	**Count**	**182**	**104**	**477**	**763**
	Expected Count	**182.0**	**104.0**	**477.0**	**763.0**
	Among PCR patients%	**23.9%**	**13.6%**	**62.5%**	**100.0%**
	Among same occupation%	**100.0%**	**100.0%**	**100.0%**	**100.0%**

The given table presents a statistical analysis of the distribution of PCR test results among individuals of different occupations, namely Nurse/Caretaker, Doctor, and Other. Among the total sample of 763 individuals, 197 people tested positive for PCR, and 566 tested negative. The table highlights that the highest percentage of positive PCR test results belonged to the "Other" occupation category, with 58.9% of positive results. Nurse/Caretakers and Doctors accounted for 26.9% and 14.2% of positive results, respectively. Similarly, the highest percentage of negative PCR test results belonged to the "Other" category, with 63.8% of negative results. Nurse/Caretakers and Doctors accounted for 22.8% and 13.4% of negative results, respectively. Interestingly, when analyzing the distribution of PCR test results within each occupation category, Nurse/Caretakers had the highest percentage of positive PCR tests (29.1%), followed by doctors (26.9%) and Others (24.3%). Moreover, the percentage of negative PCR test results was highest for the "Other" occupation category (75.7%), followed by doctors (73.1%) and Nurse/Caretakers (70.9%). These findings suggest a possible association between occupation and the likelihood of testing positive or negative for PCR.

Table 4. Contingency Table of PCR Positivity and Gender

		GENDER		TOTAL
		MALE	FEMALE	
PCR- Positive	**Count**	**68**	**129**	**197**
	Expected Count	**66.9**	**130.1**	**197.0**
	Among PCR patients%	**34.5%**	**65.5%**	**100.0%**
	Among both genders%	**26.3%**	**25.6%**	**25.8%**
Negative	**Count**	**191**	**375**	**566**
	Expected Count	**192.1**	**373.9**	**566.0**
	Among PCR patients%	**33.7%**	**66.3%**	**100.0%**
	Among both genders%	**73.7%**	**74.4%**	**74.2%**
Total	**Count**	**259**	**504**	**763**
	Expected Count	**259.0**	**504.0**	**763.0**
	Among PCR patients%	**33.9%**	**66.1%**	**100.0%**
	Among both genders%	**100.0%**	**100.0%**	**100.0%**

This table provides information on the distribution of PCR-positive and PCR-negative cases among males and females. Out of a total of 763 individuals, 259 (33.9%) were PCR positive, while 504 (66.1%) were PCR negative. The proportion of PCR positive cases among both genders is similar (26.3% for males and 25.6% for females), as is the proportion of PCR negative cases (73.7% for males and 74.4% for females). The expected counts of PCR positive and PCR negative cases for each gender are very close to the actual counts, indicating a good fit of the data to the expected distribution. Overall, there does not appear to be a significant difference in the proportion of PCR positive and PCR negative cases between males and females.

CHAPTER 5

5.1 DISCUSSION

Since its discovery in late 2019, the SARS-CoV-2 virus, which causes the disease COVID-19, has had a tremendous influence on the world. It resulted in a worldwide pandemic that afflicted millions of people, killed hundreds of thousands, and caused enormous economic and social damage. SARS-first CoV-2's and most visible impact has been on public health. The virus is very infectious and can lead to severe illness, particularly in elderly people and those with pre-existing medical issues. It has put a strain on healthcare systems, resulting in a scarcity of hospital beds, medical supplies, and healthcare professionals. The pandemic has also exposed disparities in healthcare access, as the virus has overwhelmingly afflicted marginalized groups.(CDC, 2020; Therese et al., 2020)

SARS-CoV-2 has also had a substantial impact on the world's economy. Many businesses have been devastated by the massive lockdowns and travel restrictions implemented to slow the spread of the virus, resulting in severe job losses and economic turmoil. The pandemic has also highlighted the vulnerability of global supply networks, resulting in scarcity of key items and substantial shifts in consumer behavior. The pandemic has also had a significant influence on the education sector. Schools and institutions have been forced to shut their doors, resulting in a move toward open and distance learning and a rise in communications technology. While this has enabled students to continue learning, it has also exposed the digital gap and perceived barriers to access technology and resources.

According to the **Table.4**, 197 (25.8%) of the 763 healthcare workers polled tested positive for SARS-CoV-2. There were 68 (34.5%) male cases and 129 (65.5%) female cases among the positive cases. The expected counts are 66.9 for males and 130.1 for females and that is to know if there is correlation between PCR results and gender. The expected counts for positive PCR tests are 66.9 for males and 130.1 for females, which are close to the observed counts of 68 and 129, respectively. Similarly, the expected counts for negative PCR tests are 192.1 for males and 373.9 for females, which are also close to the observed counts of 191 and 375, respectively. These results suggest that there is no significant association between PCR test results and gender, as the observed counts are similar to the expected counts. It also shows that male healthcare workers may have a somewhat greater proportion of positive cases than we would expect based on chance alone.

When the percentages within each category are considered, we can find that 26.3% of male healthcare professionals tested positive for SARS-CoV-2, whereas 25.6% of female healthcare workers tested positive. These percentages are similar, implying that there is no strong link between gender and SARS-CoV-2 PCR results with the p-value of 0.663.

Among all healthcare workers who tested positive, 34.5% were male and 65.5% were female. This suggests that there may be a higher proportion of positive cases among female healthcare workers. There are indications that female healthcare workers (HCWs) are possibly more vulnerable to SARS-CoV-2 infection than their male colleagues, which have been controversial but as seen in Table 3 of Chapter 4, there is no significant relationship between gender and COVID-19 infection.

Although numerous studies have revealed that female HCWs are more probable than male HCWs to catch SARS-CoV-2. For example, a study carried out in the United States discovered that female healthcare workers were more likely than male healthcare workers to test positive for SARS-CoV-2, even after controlling for variations in age, race/ethnicity, and occupation.(Dyal et al., 2020) Another Italian study discovered that female healthcare professionals were at a heightened hazard of SARS-CoV-2 infection than males, while the authors acknowledge that this outcome could be associated with variations in the duties done by male and female healthcare workers it is not yet verified that there is correlation between the two variables.(Cw et al., 2019a)

SARS-CoV-2 positivity amongst healthcare workers (HCWs) has been an ongoing source of worry since the outbreak. The issue differs significantly between countries, relying on variables such as the accessibility of personal protective equipment (PPE), the amount of communal spread, and commitment to infection methods of control.

Many countries reported greater percentages of SARS-CoV-2 positivity among HCWs in the beginning phases of the outbreak, with some findings showing prevalence rates as high as 30-40%. Nonetheless, as the pandemic proceeded and infection control techniques improved, the rates fell in general. Because of disparities in testing methodologies, reporting mechanisms, and demographics studied, comparing SARS-CoV-2 positivity among HCWs across countries can be difficult.(M. Liu et al., 2020) Other research, however, have sought to make these parallels. A study published in The Lancet in January 2021, for illustration, discovered that HCWs in Spain were at a greater risk of SARS-CoV-2 infection than the general public, with a positivity rate of 19.8% among HCWs compared to 10.2% in the general public.(Gonzalez & Solchaga, 2020) In contrast, a September 2020 research article appearing in the Journal of Hospital Infection reported that the positive rate among HCWs in Singapore was only 0.49%.(Heireman et al., 2020) Another research study released by the Journal of Hospital Infection in January 2021 compared SARS-CoV-2 positivity rates among HCWs in six countries (China, Italy, Spain, the United Kingdom, the United States, and Canada) and found that Italy (15.6%) and the United States (13.7%) had the highest rates, while China (1.6%) and Canada (2.8%) had the lowest rates.(Chou et al., 2020)

Many elements can be attributed to the reduced SARS-CoV-2 positive rates among healthcare workers (HCWs) in Singapore, China, and Canada when compared to Italy and Spain.(Chou et al., 2020) These elements are as follows:

- Infection control measures were established early in the pandemic in Singapore, China, and Canada, including routine health screening, utilizing personal protective equipment (PPE), and stringent compliance to infection control standards.
- Singapore, China, and Canada have robust public health systems, and healthcare organizations in these countries have expertise treating infectious illness outbreaks such as SARS and H1N1. This experience most likely led to the pandemic's swift response and the deployment of efficient infection control measures.

- Community transmission of SARS-CoV-2 was comparatively low in Singapore, China, and Canada during the early stages of the pandemic, compared to Italy and Spain, potentially having lowered the possibility of exposure and transmission among healthcare workers.
- Sufficient PPE availability: Healthcare institutions in Singapore, China, and Canada have ample supply of PPE, allowing for optimal PPE use by healthcare personnel and possibly lowering the risk of SARS-CoV-2 transmission.
- Comprehensive testing strategy: Singapore, China, and Canada established an extensive testing strategy early in the pandemic, allowing for the detection and quarantine of infected patients, which include asymptomatic or pre-symptomatic healthcare workers, which probably led to lessened SARS-CoV-2 positivity rates among healthcare workers.

As compared to Italy and Spain,(Chou et al., 2020) that had several factors that may have contributed to the higher rates, including:

- Increased community transmission: Italy was one of the pandemic's first and hardest-hit countries, and the country experienced a significant spike in SARS-CoV-2 community transmission. Because of the high community transmission rate, healthcare professionals are more likely to be exposed and transmitted.
- Inadequate PPE availability: There were complaints of PPE shortages in Italy and Spain during the early stages of the pandemic, which may have raised the potential of SARS-CoV-2 transmission among healthcare personnel. Inadequate PPE may also have resulted in reduced compliance to infection control practices among healthcare staff.
- Overcrowding in hospitals and healthcare facilities: The significant number of COVID-19 cases in Italy and Spain led to congestion in hospitals and healthcare facilities, potentially increasing the risk of SARS-CoV-2 transmission among healthcare workers.
- Delayed deployment of infection control measures: Italy was one of the first nations outside of China to suffer a substantial outbreak of SARS-CoV-2, and the country experienced difficulty in adequately responding to the pandemic. The

delayed adoption of infection control measures, such as regular health screening, the use of suitable PPE, and rigid adherence to infection prevention and control regulations, may have led to a greater rate of SARS-CoV-2 positive among healthcare personnel.

- Limited testing capacity: In the early stages of the pandemic, Italy and Spain had limited testing capacity, which may have led to a decreased identification of asymptomatic or pre-symptomatic healthcare workers infected with SARS-CoV-2, potentially contributing to even further transmission among healthcare professionals.

Healthcare workers (HCWs) are crucial in combating the continuing SARS-CoV-2 pandemic. Because of their close interactions with infected patients and regular use of personal protective equipment, (PPE)they are at a higher risk of viral exposure. HCWs have a higher chance of catching SARS-CoV-2 than the general population, according to Section 2.20 where this study emphasized more on. This heightened risk can result not only in illness and absence from work, but also in staffing issues and delays in healthcare delivery. Additionally, the psychological burden of working throughout a pandemic cannot be ignored. Because of the nature of their profession and the dread of infecting themselves and their nearest and dearest, HCWs are experiencing unprecedented levels of stress, anxiety, and burnout.(Lai et al., 2020)

Although there is no statistical relationship between occupation and SARS-CoV-2 in our study, we saw that the healthcare workers with the highest contraction of the disease were nurses due to their close and frequent interaction with the infected patients and also duration of shift in the hospital which is longer than others in the healthcare sector. Followed by nurses were patient counselors, for obvious reasons because they assist patients with symptom management, coping skills, and connecting with safety nets. They can also assist patients in processing their pandemic-related perceptions and emotions, as well as making knowledgeable choices about their mental wellbeing. Furthermore, patient counselors can assist healthcare workers and other critical employees who are on the forefront lines of the pandemic. As a result of their exposure to the virus and its consequences, these professionals may endure heightened stress, burnout, and trauma. Patient counselors can assist these employees in managing their psychological and emotional health and maintaining their well-being as they continue to work.(Lai et al., 2020)

Since it can amplify even extremely minimal quantities of viral genetic material, PCR (polymerase chain reaction) is much more precise compared to other techniques for identifying SARS-CoV-2 (the virus responsible for COVID-19) and it's the reason why it is used to test for SARS-CoV-2 in HCWs in this study. This means that PCR testing can identify the existence of the virus in a specimen regardless of whether the viral load is very minimal, which can be highly pertinent in early or asymptomatic infections where viral load may be lesser.(Perchetti et al., 2021; Tang et al., 2020)

PCR utilizes a particular set of primers (short strands of DNA) that are tailored to attach to viral genetic material in the sample if it is existent. The primers then start a chain of enzyme reactions that multiplie the viral genetic material which makes SARS-CoV-2 detection easier. Antigen tests, for example, are much less sensitive than PCR and it may yield false negatives in circumstances where the virus load is minimal. Antigen tests, on the other hand, are frequently quicker and less costly than PCR tests, which makes them appropriate in instances where quick testing is required.(Perkmann et al., 2020; Tang et al., 2020)

Because of its excellent sensitivity and specificity, PCR is still the preferred method for SARS-CoV-2 diagnosis. Since it can amplify tiny quantities of genetic material from the SARS-CoV-2 virus to trace amounts, PCR (polymerase chain reaction) is very sensitive and selective. PCR employs a particular set of primers and probes to target a particular portion of the viral genome. Whenever a virus-containing sample is introduced to the PCR reaction, the primers and probes attach to the viral genetic material, causing amplification of that portion of the genome to begin. The number of replicates of viral genetic material in the sample grows mountingly as the reaction progresses, allowing even trace levels of the virus to be recognized. Furthermore, PCR can distinguish between various viruses of different kinds depending on the sequence of their genetic material, which makes it extremely specific.(Perchetti et al., 2021)

PCR can also be programmed to target numerous sections of the virus genome, boosting its sensitivity and specificity. This indicates that even in samples with very low viral loads, PCR testing can confirm the existence of the SARS-CoV-2 virus.

For the collection of nasal swab samples for PCR testing which is a standard method for identifying respiratory viruses, particularly COVID-19. The following are the steps involved in collecting nose swab samples for PCR:

When doing the swab collection, the healthcare provider should wear proper personal protective equipment (PPE), such as gloves, gowns, and face shields. The patient should be told of the process and urged to slightly lean their head back.(M. Liu et al., 2020)

For nasal swab collection, a soft, sterile, elastic swab with a plastic tip, such as a nylon flocked swab, is commonly utilized. The swab should be carefully inserted into the patient's nostril and aimed directly back along the base of the nose, parallel to the palate, until resistance is met. If the swab is forced, the patient may feel mild discomfort or a feeling of swelling in the nose. To gather enough material, the swab should be spun multiple times while in contact with the nasal mucosa for 15 seconds maximum. After that, gently remove the swab and place it in a sterile tube containing viral transport medium. The same method should be repeated for the other nostril, if necessary, with a separate swab. The collected sample should be taken to the laboratory as soon as possible, ideally within 24 hours of collection, for PCR testing. The sample must be kept at 2-8°C until it is transferred to the lab.(Perchetti et al., 2021; Perkmann et al., 2020; Tang et al., 2020; Zhang et al., 2020)

Sample Collection: The first step is to extract the sample containing the DNA of interest by PCR sample collection. The sample could be anything, such as blood, saliva, tissue, or any other biological material containing DNA. Based on the source, multiple approaches may be required to obtain the sample. Once the sample has been obtained, it must be preserved until the DNA can be retrieved. The material type taken will determine the technique of preservation. Blood samples, for example, may require storage in an anticoagulant solution, whereas tissue samples may require freezing or storage in a preservative solution.(Perchetti et al., 2021; Tang et al., 2020; Wang et al., 2020)

There are several techniques for extracting DNA, including using commercial kits or performing hand extraction with chemicals. It is critical to quantify the amount of DNA present in the sample after it has been extracted. This can be accomplished with a spectrophotometer or through other means. After quantifying the amount of DNA, the sample may need to be diluted to a specified concentration to guarantee optimal PCR performance. The amount and quality of the extracted DNA will determine the suitable dilution. Lastly, the extracted and diluted DNA is added to the PCR mix, which comprises the essential parts for PCR amplification, including Taq polymerase, primers, and nucleotides. After collecting the sample, it is lysed to release the RNA. Lysis can be accomplished through physical, chemical, or mechanical means. Sonication, crushing with a pestle and mortar, or employing detergents to break apart cell membranes, for example, can all be used to disturb cells or tissue.(Azzi et al., 2020; Perchetti et al., 2021; Wang et al., 2020)

RNA is extremely fragile and easily degraded by ribonucleases, which are enzymes that break down RNA. As a result, it is critical to stabilize the RNA after lysis. This is accomplished by including an RNA stabilization reagent, which deactivates the ribonucleases while preserving the RNA. After stabilizing the RNA, it must be separated from other biological components. There are several methods available for RNA purification, including column-based purification, precipitation, or using magnetic beads. These approaches rely on the idea of RNA attaching to a specific matrix, whereas other biological components do not. After purification, the amount of RNA must be determined. This can be accomplished with a spectrophotometer or through other methods such as gel electrophoresis or qPCR. In addition to quantifying, the quality of the RNA must be assessed. This can be accomplished using a Bioanalyzer or other means. The quality of RNA can impact the success of downstream applications such as PCR. Because PCR requires DNA as a template, RNA must be transformed to cDNA (complementary DNA) via a method named as reverse transcription. This is accomplished by the use of reverse transcriptase, which generates a complementary DNA strand from the RNA template.(Perchetti et al., 2021; Tang et al., 2020)

The next step is to perform reverse transcription mix containing the RNA template, reverse transcriptase enzyme, oligo(dT) primer, dNTPs, and buffer is produced to execute reverse transcription. The oligo(dT) primer heat and cool (anneals) to the poly(A) tail of the mRNA, allowing reverse transcriptase to begin cDNA synthesis. In order for the reverse transcriptase enzyme to function, the RNA template is incubated with the reverse transcription mix at an appropriate temperature. The temperature and time of the reaction are determined by the type of reverse transcriptase enzyme utilized and the technique.(Tang et al., 2020)

Following the completion of the reverse transcription reaction, the cDNA must be purified from other reaction components such as residual RNA or primers. This can be accomplished through a variety of means. Such methods include column-based purification, precipitation, and the use of magnetic beads. Following purification, the amount of cDNA must be measured with a spectrophotometer or another method, such as gel electrophoresis or qPCR. In relation to quantification, the quality of the cDNA must be evaluated. This can be accomplished through the use of a Bioanalyzer or other means. The efficacy of downstream applications such as PCR might be influenced by the quality of cDNA.(Azzi et al., 2020; Perchetti et al., 2021; Zhang et al., 2020)

Upon the transformation of isolated RNA to complementary DNA (cDNA), the DNA template, primers, probe, Taq polymerase enzyme, dNTPs, and buffer are combined to make a real-time PCR mix. The probe is fluorescently tagged with a reporter dye and a quencher dye. When excited, the reporter dye gives a fluorescent signal, but the quencher dye precludes detection until the probe is split during the PCR reaction. The real-time PCR mix is poured into PCR plate wells with negative and positive control wells. To avoid evaporating, the dish is then covered with a lid. The PCR plate is then placed in a real-time PCR apparatus, which can detect the fluorescence of the reporter dye in the course of the PCR reaction. The instrument is calibrated with the proper thermal cycling protocol, which includes an initial denaturation phase, followed by a predetermined number of amplification cycles, each of which includes a denaturation, annealing, and extension step.(Perkmann et al., 2020; Tang et al., 2020)

To thoroughly denature the double-stranded DNA template and trigger the Taq polymerase enzyme, the PCR reaction starts with a high-temperature denaturation step, usually at 95°C for 10 minutes.(Zhang et al., 2020) Then, the fluorescently tagged/labeled probe attaches to the target DNA sequence and is split by the Taq polymerase enzyme during the annealing and extension steps of the PCR reaction, releasing the reporter dye and enabling it to generate a fluorescent signal.(Perkmann et al., 2020) The light produced is correspondent to the number of DNA amplified during the PCR reaction. The fluorescence produced by the reporter dye is detected by the real-time PCR apparatus at every cycle of the amplification process. The light produced is proportionate to the quantity of DNA increased during the PCR reaction. The threshold cycle (Ct) value is the cycle where the fluorescence signal hits a certain threshold and can be used to quantify and evaluate the quantity of DNA in multiple samples.(Tang et al., 2020)

Following the completion of the cycling process, a melting curve analysis can be done to confirm the particularity of the PCR reaction and to detect any nonspecific amplification products. The temperature is gradually raised, melting the double-stranded DNA and decreasing the fluorescent signal. The melting temperature (Tm) of the amplified DNA product is the temperature at which the fluorescent signal drops. The next step is analyzing PCR data which is determining the threshold cycle (Ct), i.e the number of cycles at which the fluorescent signal of the amplified DNA exceeds a predefined threshold. Ct values are an indirect indication of the quantity of target DNA in a sample, with lower Ct values corresponding to more target DNA.(Zhang et al., 2020)

Then, a standard curve is created by graphing the Ct values against the logarithm of the target DNA concentrations using known concentrations of the target DNA. The slope of the standard curve is used to calculate the PCR reaction effectiveness, which signifies the amount of target DNA is multiplied in each PCR cycle.(Tang et al., 2020)

Which leads to the next step, absolute quantification, which compares the Ct values of the sample to the standard curve to determine the precise amount of target DNA in a sample. By interpolating/interjecting the sample's Ct value on the standard curve and applying the equation that connects the number of target DNA to the Ct value, the precise amount of target DNA may be estimated.(Perkmann et al., 2020)

Following absolute quantification, comes relative quantification which compares the amount of target DNA present in multiple samples. This can be done by evaluating the Ct values of the target DNA in each sample, with a higher Ct value indicating less target DNA. Melting curve analysis is utilized to ensure that the PCR reaction is specific and that the expected amplicon is present. Gradually increasing the temperature leads the DNA to denature and the fluorescent signal to drop. The melting temperature (Tm) of the amplified DNA product can be evaluated to the predicted Tm to ensure the specificity of the PCR reaction.(Tang et al., 2020)

The last step is data normalization which is a method of correcting for differences in sample preparation, DNA quality, and PCR effectiveness. This is accomplished by employing an internal control, such as a housekeeping gene, to equalize the target DNA Ct values to the internal control's Ct values and calculating the target gene's relative expression level.(Wang et al., 2020)

The Ct value regarded positive for SARS-CoV-2 identification may also be affected by the type of sample analyzed. A Ct value of 40, for example, is deemed positive for nasal swab samples, whereas a Ct value of 37 is considered positive for saliva samples.(Perchetti et al., 2021)

It is critical to remember that the Ct value should be evaluated in the context of the patient's clinical presentation and epidemiological background being tested. A positive result does not always signify active infection or infectiousness, and a negative result does not exclude the potential of infection totally. It is critical to follow up with healthcare providers to ensure proper diagnosis and treatment.(Tang et al., 2020)

5.2 CONCLUSION

In conclusion, the study aimed to quantify SARS-CoV-2 titers among healthcare workers (HCWs) near the conclusion of the first wave of the pandemic, determine likely groups with higher infection rates, and identify probable infections sources both within and outside of the professional setting. The study's findings provide significant input into the virus's proliferation and transmission among HCWs and may assist future attempts to regulate and avoid the spread of COVID-19. The findings emphasize the importance of sustained monitoring and the implementation of efficient HCW protection measures, such as regular testing, proper utilization of personal protective equipment, and universal immunization. Overall, the study emphasizes the crucial significance of HCWs in the battle against COVID-19 and the necessity for additional research. Because of their increasing exposure to infected patients, healthcare workers (HCWs) continue to face a high risk from the SARS-CoV-2 virus. Healthcare workers (HCWs) have been at the forefront of the COVID-19 pandemic since its onset. They have played a crucial role in diagnosing, treating, and caring for patients with COVID-19, putting themselves at increased risk of exposure to SARS-CoV-2 both inside and outside of the professional setting. HCWs are at risk of SARS-CoV-2 exposure in the workplace due to interaction with COVID-19 positive patients or those with COVID-19 symptoms.(Lai et al., 2020) Failure to apply adequate infection prevention and control measures, such as PPE, hand hygiene, and environmental cleaning and disinfection, can further increase the risk of transmission.(Gershon et al., 1995) Working in high-risk locations such as critical care units, emergency departments, or COVID-19 certified units, as well as exposure to contaminated surfaces, equipment, or materials in healthcare institutions, are all possible sources of infection for HCWs.(Lai et al., 2020)

HCWs are at risk of SARS-CoV-2 exposure outside of the workplace due to close interaction with family members or household members who have COVID-19 or COVID-19 symptoms.(Gershon et al., 1995) Participation at large meetings or events when physical separation procedures are not in place or are not followed can further increase the likelihood of transmission. Travel to locations with high COVID-19 transmission rates, or contact with people from these areas, can potentially raise the risk of infection. Not adhering to public health norms in the community, such as the usage of face coverings, physical distance, and hand hygiene, can further raise the likelihood of transmission for HCWs.(Cw et al., 2019b)

To limit the risk of SARS-CoV-2 transmission, HCWs must be diligent in infection control and prevention efforts within as well as outside the professional setting.

Following adequate PPE practices, maintaining excellent hand hygiene, sticking to physical distancing procedures, and avoiding high-risk circumstances or exposures are all part of this. HCWs must also take care of their physical and emotional health, which includes obtaining appropriate rest, fitness, and diet, as well as seeking help and assistance when necessary.(Gershon et al., 1995)

5.3 RECOMMENDATION

Given the potential sources of infection for healthcare workers (HCWs) inside and outside of the professional setting in the context of SARS-CoV-2, it is recommended that healthcare organizations implement methods to mitigate the likelihood of COVID-19 spread at work. This can include providing personal protective equipment (PPE), regular testing and training on infection control practices, and ensuring a safe working environment for HCWs.(CDC, 2020) As new information comes to light, this training should be revised. Second, healthcare institutions should ensure that suitable PPE is readily available to HCWs. This includes assuring an adequate supply of personal protective equipment (PPE), suitable fit testing, and correct wearing and removing procedures.(M. Liu et al., 2020) It is important for HCWs to continue taking actions to safeguard themselves and their patients against COVID-19, including getting vaccinated and adhering to guidelines on PPE and infection control.(Gershon et al., 1995)

Third, healthcare institutions should emphasize HCW protection by instituting appropriate administrative controls, such as grouping COVID-19 positive patients, restricting visitation, and conduct periodic screening and diagnostics of HCWs. Fourth, public health officials should continue to convey concise and consistent information to the general population about infection control and prevention strategies, such as the use of face coverings, physical distance, and hand hygiene.(Chou et al., 2020) Lastly, healthcare facilities should take into account HCWs' physical and mental well-being as a priority by offering tools and support for dealing with the pandemic's mental and emotional devastating impact. This includes having access to psychological health resources, psychological counseling, and relaxation and self-care options.(M. Liu et al., 2020)

Adopting these guidelines will assist healthcare institutions and public health officials minimize the likelihood of SARS-CoV-2 transmission to HCWs both inside and outside of the professional setting, all while aiding HCWs' physical and mental well-being during the difficult time.(Gonzalez & Solchaga, 2020) The hazard of nosocomial infection can be evaluated in two instances: daily risk of infection and overall amount of time of exposure. Patients and healthcare workers who spend a brief time in the high daily risk department share the same chance of being infected as patients and healthcare workers who stay longer in the low daily risk sector.(M. Liu et al., 2020) Every department must minimize daily risk by several protective and sanitation measures, as well as shorten patient care time by eliminating superfluous treatment processes.(Cw et al., 2019b)

Discharged patients' wards must be thoroughly disinfected. All surfaces, including bedding, curtains, vents, and lighting, should be thoroughly cleaned. Adequate air isolation measures, such as individual rooms, negative pressure, regular airflow, and electric air-purifying respirators are required for medical procedures that may produce aerosols. Every department should create its disinfection approach, making sure that everywhere is sanitized and treated at the end of each day's activity. (Chou et al., 2020)

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