Nanoparticle against SARs-CoV-19: Applications for prevention, diagnosis, and treatment of COVID-19 and future perspectives

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ABSTRACT

The catastrophic COVID-19 pandemic, which was caused by the recent SARS-CoV-2 epidemic, had a significant effect on humanity and the global healthcare system. SARS-CoV-2 mostly spreads by symptomatic carriers who then enter the host cell through the ACE2 and TMPSSR2 receptors and cause organ damage. Standard diagnostic procedures and treatment approaches are not as effective as they need to be to stop SARS-CoV-2 from spreading. The SARS-CoV-2 coronavirus, which first demonstrated human-to-human transmission in December 2019 and has infected millions of individuals in only a few months across 213 different nations, is the pathogen that causes this sickness. Its potential to spread through asymptomatic carriers has greatly strained the diagnostic resources that are now available. COVID-19 vaccines are still in the development stage, and there are currently no clinically shown treatment approaches that block the effects of this virus. Expanding testing capabilities, creating efficient medicines, and creating secure vaccinations that offer long-lasting immunity are all strategies that need to be investigated. Numerous medical applications, including biosensing, drug delivery, and antimicrobial therapy, use nanoparticles (NPs) extensively. SARS-CoV-2 is an enveloped virus with a diameter of 60–140 nm and particle-like properties. Due to their physical similarity and ability to closely imitate the virus and bind strongly with its proteins. Therefore, NP-based approaches to combating this virus have enormous potential. NPs had previously been discovered to be powerful weapons against several viruses, particularly those belonging to the Coronaviridae family. This review will provide a one-site informative platform for researchers to explore the crucial role of nanoparticles in managing the COVID-19 curse more effectively.

Keywords: *Nanoparticles, SARS-CoV-2, lipid nanoparticles, silver nanoparticles, NPs vaccines.*

1. INTRODUCTION

Human coronaviruses are members of the family *Coronaviridae*, order *Nidovirales*, suborder *Cornidovirineae*, and subfamily *Orthocoronavirinae*, the latter of which has four genera known as alpha, beta, gamma, and delta CoVs. Viruses that affect both the upper and lower respiratory tracts are included in the category of human coronaviruses. Coronaviruses that cause upper respiratory tract infections (URTIs) include HCoV-229E

(*Alphacoronavirus*, subgenus *Duvinacovirus*), HCoV-NL63 (*Alphacoronavirus*, subgenus *Setracovirus*), HCoV-OC43, and HCoV-KHU1, the latter two being related to *Betacoronavirus* subgenus *Embecovirus*. They account for 10%–30% of cold infections in people. SARS-CoV, SARS-CoV-2, and MERS-CoV (*Betacoronavirus* subgenus, *Merbecovirus*) are the three coronaviruses that cause severe lower respiratory tract infections (LRTIs)[1].

SARS-Co-2, the coronavirus that causes severe acute respiratory syndrome, is the cause of the 2019 coronavirus infection illness (COVID-19). It is one of the infectious illnesses with the fastest global spread and highest mortality rate. Since December 2019, SARS-Co-2 has spread rapidly over the world, killing thousands of people. The COVID-19 virus, also known as SARS-CoV-2, was first discovered in a bat in Wuhan, China, in December 2019. The WHO labeled the viral outbreak, a health emergency in January 2020. A total of 40 distinct SARS-CoV-2 strains have been identified in recent years. There is currently no acceptable or efficient therapy for the illness. Drug resistance and non-specific toxicity are problems with conventional antiviral medications. Due to its straightforward engineering and capacity to target structural viral proteins inducing the creation of antibodies that neutralize the virus, RNA-based vaccinations have emerged as potential alternatives. However, they are linked to some undesirable immune responses, necessitating the continued development of more advanced, site-specific, and secure antiviral medicines, vaccines, and agents[2].

Around the nation, this epidemic has caused uncomfortable situations that have prompted rules like social distancing, lockdowns, and other measures. A report from Guangdong, China, in November 2002 said that SARS was the first sickness the world had to deal with in the new millennium. SARS, Ebola, Swine Flu, and Middle East Respiratory Syndrome don't seem to be able to equal the number of deaths brought on by COVID-19. Over 4 million people have died from COVID-19 infections, which have affected over 189 million people globally. An outbreak has been swiftly transformed into a pandemic by COVID-19[3].

1.1.A brief outlook of SARS-CoV-2: molecular structure: A single-stranded RNA virus containing active-sense single-stranded RNA, SARS-CoV-2 has a crown-like form. Its diameter ranges from 600 to 1400 A^o and is a *Coronaviridae* family member. Particles that are extracellular and free may be examined with an electron microscope. The pathogenicity of the virus is caused by its spike (S), envelope (E), membrane (M), and nucleocapsid (N) components. (Figure 1)[3]**.**

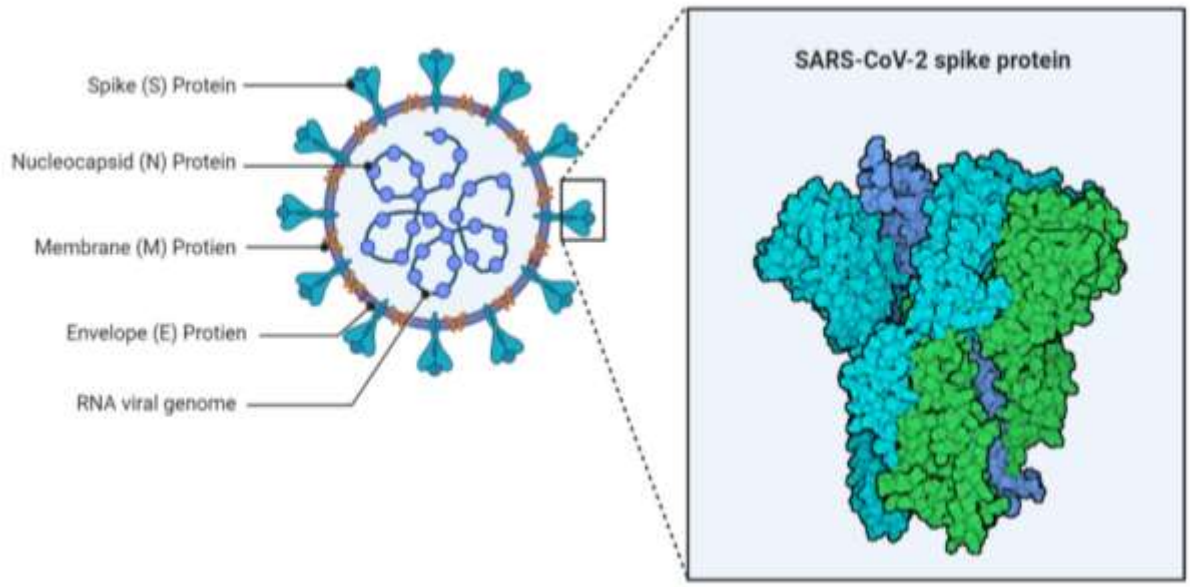

Figure. 1 Structure of COVID-19

The transmembrane protein known as the spike, or S glycoprotein, has a molecular weight of around 150 kDa and is located in the virus's outer layer. By attracting the angiotensin-converting enzyme 2 (ACE2) produced in lower respiratory tract cells, the S protein creates homotrimers that protrude from the viral surface and aids in the attachment of envelope viruses to host cells. The host cell's furin-like protease splits this glycoprotein into the S1

and S2 subunits. With the receptor binding domain makeup, part S1 determines the host-virus range and cellular tropism, while part S2 mediates viral fusion in transmitting host cells.

The structural element of CoV that is structurally attached to the virus's nucleic acid is the nucleocapsid, also known as the N protein, which is found in the endoplasmic Reticulum-Golgi area. The protein is implicated in activities about the viral genome, the viral replication cycle, and the biological response of host cells to viral infections because the protein is attached to RNA. Additionally, the N protein is highly phosphorylated, which may alter its structural makeup and increase its affinity for viral RNA.

The membrane protein, also known as the M protein, is another crucial component of this virus. It is the most structurally complex protein and affects how the virus's envelope is shaped. All other structural proteins can bind to this protein. By stabilizing the N protein-RNA complex inside the internal virion, binding with M protein aids in the stabilization of nucleocapsids or N proteins and facilitates the completion of viral assembly. The final part of the SARS-CoV structure is the envelope, or E protein, which is the smallest protein and essential for the development and maturation of the virus.

Another essential part of this virus is the membrane protein, also called the M protein. It influences how the virus's envelope is structured and is the most structurally complicated protein. This protein is capable of binding to all other structural proteins. Binding with M protein assists in the stabilization of nucleocapsids or N proteins and makes it easier for the viral assembly process to be completed by preserving the N protein-RNA complex inside the internal virion. The envelope, or E protein, is the last component of the SARS-CoV structure and is crucial for the growth and maturity of the virus. It is the smallest protein[4].

2. ETIOPATHOGENESIS OF COVID-19 VIRUS

The processes in the pathogenesis of the COVID-19 virus's dissemination to the host body included attachment, penetration, biosynthesis, maturation, and viral release. The interaction between the viral spike protein and the host cell's ACE2 receptor causes the viral attachment. The process of direct membrane attachment or fusion with the host receptor is known as penetration and involves proteolytic cleavage at the S1/S2 border. The RNA is subsequently released into the host cell's cytoplasm, proceeds toward the nucleus, and eventually enters the nucleus, where biosynthesis occurs through transcription, translation, and protein synthesis. The newly created protein passes via the endoplasmic reticulum, where it matures and is released back into the body by the Golgi apparatus. The COVID-19 virus mostly affects the lungs and causes symptoms including a dry cough, fever, body aches, and dyspnea. The virus can reach the blood through the respiratory tract's epithelial cell layer and affect several vascular functions. As a result, histopathological investigations of the lungs of an infected patient might show alveolar destruction, hyaline membrane development, desquamation of pneumocytes, and cellular fibro myxoid transudes[3].

3. ROLE OF NANOTECHNOLOGY IN VIRAL DISEASE

Through numerous elements including targeted drug delivery, drug development possibilities, etc., nanotechnology and material science have already demonstrated their value in supporting the traditional medicinal chemistry process. Preventing contamination is the most efficient way to treat viral illnesses, thus using nanomaterials appropriately may be advantageous in this aspect. According to Van Doremalen *et al.*, compared to surfaces made of plastic and steel, copper had the lowest survival rate for SARS-CoV-2. The vitality of the virus will considerably decline with an increase in the copper concentration of alloys, according to further research that also supported this conclusion. This characteristic applies to other coronaviruses that have been described, and it was related to the production of reactive oxygen species (ROS) by copper nanoparticles. Therefore, it becomes urgently important to pay attention to the antibacterial and antiviral capabilities of nanoparticles, which are frequently described in scientific literature. Quantum dots, nanotubes, metal oxide nanoparticles, polymer nanocomposites, two-dimensional nanomaterials, lipid nanoparticles, and other types of nanomaterials were described for their strong antibacterial activity and therefore entered the platform of COVID-19 control[5].

4. CURRENT METHODS OF COV-19 DIAGNOSIS

Diagnostic techniques that detect viral nucleic acids, viral antigens, or serological tests are required to confirm SARS-CoV-2 infection. fast antigenic and fast antibody tests are easier to do, cost less money, and take less time to complete. They also don't require highly experienced personnel. The indirect enzyme-linked immunosorbent assay (ELISA) methodology is the foundation for the majority of COVID-19 immuno enzymatic serological assays.

With this technique, researchers can gather extremely sensitive and precise data in a brief amount of time (between 1 and 3 hours**).** The effectiveness and specificity of several alternative techniques for diagnosing COVID-19 infection, including viral culture and electron microscopy, NGS, clinical investigations and imaging techniques (CT Scan), biosensor COVID-19 testing techniques, loop-mediated isothermal amplification (LAMP), CRISPR/Cas-based COVID-19 testing methods, and digital PCR COVID-19 testing methods, are still debatable[6].

5. USAGE OF NANOPARTICLES IN THE DETECTION OF COV 19

SARS-CoV-2 diagnosis is a crucial first step that depends on knowledge of the genetic and structural proteins that make up the virus. Upon entrance, the SARS-CoV-2 virus specifically binds to the ACE2 protein on host cells to begin replicating. The C-reactive protein and serological components of the current SARS-CoV-2 diagnostic method are dependent on their existence. C- reactive protein detects the presence of viral particles by interacting with a portion of the SARS-CoV-2 genetic code, whereas the serological approach depends on the intensity of the immune response to the virus[7].

Today, a variety of techniques are used as highly sensitive diagnostic systems to identify SARS-CoV-2, and numerous new techniques still need to be researched. To increase the sensitivity and reliability of the tests, US researchers created a quick test utilizing gold nanoparticles. They described it as an effective and quick method for the diagnosis of COVID-19. The authors plan to link a particular molecule to the gold nanoparticles to make them identify a specific protein from the genetic makeup of the SARSCoV-2 virus[8].

The biosensor and the virus' gene sequence interacted, and the gold nanoparticles are what caused the liquid reagent to change color from purple to blue. In this manner, a direct diagnosis of the SARS-CoV-2 virus in the sample was revealed by a visual change. They facilitate the target detection of the virus by enabling the linking of biological molecules to create hybrid biological Au nanoparticle structures. A diagnostic system based on Bio-Au nanoparticles exhibits improved test sensitivity and a wider detection range, which shortens the detection time[9]. A quick detection assay for IgG and IgM antibodies against SARS-CoV-2 has also been developed by researchers using gold nanoparticles. This kit helps to shorten the diagnostic process and displays the results in less than 10 minutes[10].

By extending the sample life, several of the issues with the current diagnostic method can be reduced. Due to the lack of refrigerated transportation, samples taken from patients must be examined every once to prevent the deterioration of components like RNA and protein. In this regard, researchers have produced an incredible amount of work to create a sample collecting kit comprising an RNA stabilization fluid. The kit preserves viral RNA such that it can be transported safely from one location to another as needed for up to a week at room temperature. The development of a biosensor for the detection of SARS-CoV-2 uses two-dimensional nanoparticles in combination with functional DNA receptors[11]. A diagnostic method based on a nanoparticle that employed isothermal amplification via a multiplex transcription loop was created by Zhu *et al*., Their examination of 33 oropharyngeal samples from COVID-19 patients showed 100% sensitivity and 100% specificity compared to 96 samples from individuals without the disease[12]. Nanoparticles are useful in the diagnosis, and treatment of COVID-19 (Figure 2).

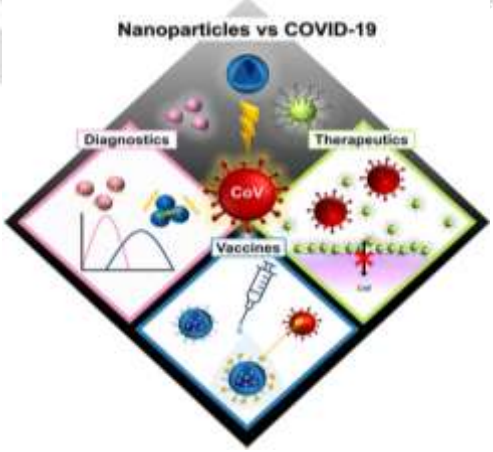

Figure 2. Nanoparticles vs COVID-19

6. NANOPARTICLE VACCINE FOR COVID-19 IN DEVELOPMENT

Along with conventional vaccination approaches (such as inactivated, live, and recombinant protein vaccines) and DNA and vector-based vaccines, nanoparticle vaccines have a singular chance to progress vaccination technology and give workable remedies to the ongoing pandemic and those in the future. In a broad sense, nanoparticles are programmable, nanoscale-sized particulate entities that resemble natural viruses in their structural characteristics. They are very promising platforms for the creation of next-generation vaccines because of their adaptive architecture, which offers paths to trigger potent nanotechnology albumin-bound responses or wider antibody-based immunity that may better account for the evolution and variety of viral infections. There are currently 60 more candidates for nanoparticle-based vaccines in various phases of preclinical development, and at least 26 of these have moved into human clinical trials. These vaccines come in a variety of forms, including micelles, protein nanoparticles, lipid nanoparticles, and virus-like particles (VLPs).

Nanoparticle vaccines may be split into two categories based on antigen loading strategies: (1) those that encapsulate vaccine antigens or nucleic acid cargos within their cores, and (2) those that display vaccine antigens on their surfaces. Nanoparticles displaying vaccine antigens can engage antigen-presenting cells (APCs) and/or effectively promote B cell receptor (BCR) cross-linking, leading to potent immunogenicity, which is a key characteristic of antigen-encapsulating nanoparticle vaccines[13].

6.1 Antigen-encapsulated nanoparticles

6.1.1. Lipid nanoparticles: To transport nucleic acid cargos (DNA and RNA) encoding vaccination antigens, lipid nanoparticles (LNPs) are used. Encapsulation within LNPs serves to both ease the cellular transport of mRNA, which is difficult owing to size and charge restrictions, and to protect mRNA cargo against fast destruction by RNases. An essential part of LNPs is an ionizable lipid, which is neutral at physiological pH but positively charged at low pH to enable the encapsulation of negatively charged mRNA[13].Through endocytosis, LNPs enter cells. The low pH of endosomes causes the ionizable lipid to become positively charged, which can rupture endosomal membranes and release the mRNA cargo for protein translation. After the availability of a viral genome, mRNA-LNPs may be completely synthesized and produced without the need for live cells, enabling the fast manufacturing of vaccines at scale within weeks[14].

The first two COVID-19 vaccines to be authorized for use were LNPs that delivered engineered S mRNA. In clinical testing, the two doses of the mRNA-LNP vaccines from Moderna and BioNTech/Pfizer both showed effectiveness against the Wuhan-Hu-1 SARS-CoV-2 of more than 90%. The BioNTech/Pfizer BNT162b2 mRNA vaccine has been used to vaccinate more than 80% of the adult population with at least one dose in situations where mass vaccination is practiced, such as in Israel. The two mRNA-LNP vaccines were 94% effective against COVID-19 hospitalization in completely immunized older persons aged 65 in the United States, too. In a current phase 1 experiment, neutralizing antibody responses following LNP vaccination are long-lasting and last for longer than six months in all healthy adult participants. The major SARS-CoV-2 VOC in circulation in several nations right now is the Delta form. When compared to the Alpha version, the Delta variant's symptomatic condition was much more resistant to a single dosage of BNT162b2 (30.7% vs. 48.7% effective). BNT162b2 was 93.7% effective against Alpha and 88% effective against Delta after two doses[13].To combat new SARS-CoV-2 mutations, the next generation of reformulated mRNA-LNP vaccines is being developed.

6.1.2. Polymer nanoparticles: In the past, nanoparticle vaccines for various coronaviruses, such as SARS-CoV, MERS-CoV, or hCoV, have frequently been made using polymers, such as chitosan, poly (lactic-co-glycolic acid) (PLGA), or polyethylene mine (PEI). Upon intranasal delivery, positively charged chitosan can electrostatically interact with negatively charged mucus sialic acid to facilitate nanoparticle attachment in airway epithelial surfaces. Mannose sugars were also used to adorn the chitosan nanoparticles' surface. Increased nanoparticle shuttle activity to the network of follicular dendritic cells and deposition in the germinal centers of lymph nodes may result from innate immune detection of mannose[15]. Although there are currently no reports on the immunogenicity of RBD-mannosylated chitosan nanoparticles, a similar approach was used to transport swine influenza A viral antigen, which resulted in strong cross-reactive IgA and IgG antibody titers and protection against a viral challenge[13].

6.2. Antigen-presenting nanoparticles

6.2.1. Virus-like particles: VLPs are nanoscale self-assembled viral structural proteins that have the potential to carry vaccination antigens. They imitate the physical and molecular characteristics of natural viral virions, but since they lack genetic material, they are not contagious and cannot reproduce. Several SARS-CoV-2 VLP vaccines are being developed, with CoVLP being the most advanced and now undergoing phase 2/3 clinical trials. CoVLP, created by Medicago, is made by temporarily transfecting tobacco plants with *Agrobacterium*[16]. Enveloped vesicles spontaneously form and exhibit surface-stabilized pre-fusion S trimers. In a phase 1 experiment, two doses of CoVLP made with AS03 adjuvant resulted in robust nanoparticle albumin-bound responses that were 10 to 50 times greater than those seen in COVID-19 convalescent subjects. In comparison to soluble antigens, a single dose of a VLP vaccination produces better quality and larger antibody responses. When compared to recombinant prefusion S, an encapsulated VLP produced from the murine leukemia virus and exhibiting prefusion S generated strong nanoparticle albumin-bound responses in mice and provided greater protection for hamsters against the contagious SARS-CoV-2 challenge[17].

6.2.2. Micillies: A prime example of a micelle-delivered SARS-CoV-2 protein is the vaccine produced by Novavax, which is presently undergoing late-stage phase 3 clinical development. Micelles are self-assembling amphiphilic structures. The amphiphilic detergent polysorbate 80 (Tween 80) is used in this vaccine (NVXCoV2373), which is also co-formulated with the saponin-based Matrix-M1 adjuvant[18]. However, the vaccination only achieved a lower overall effectiveness of 49.4% in the phase 2b study (NCT04533399) conducted in South Africa, where the Beta VOC circulates mostly. A modified vaccination that targets the Beta version is being created[19].

6.2.3. Self-assembling protein nanoparticles: Delivering SARS-CoV-2 subunit protein vaccines (S and RBD) has also been produced using protein-based nanoparticle systems. A phase I clinical study (NCT04784767) for a self-assembling SARS-CoV-2 S ferritin nanoparticle (SpFN) is presently underway. Helicobacter pylori ferritin, a common iron-storage protein with 24 subunits, was genetically fused with a gene expressing prefusion-stabilized S ectodomain to create SpFN. When SpFN is expressed in mammalian cells, it self-assembles into a nanoparticle with 8 vertices that have 3-fold symmetry, making it easier for trimeric S proteins to be shown in an organized fashion. In preclinical testing, the administration of two doses of 50 mg SpFN co-formulated with a liposomal adjuvant to rhesus macaques resulted in strong nanoparticle albumin-bound titers and protected the animals from intratracheal and intranasal SARS-CoV-2 challenge. Lower and upper airway viral replication as well as diminished pulmonary pathology were noted[13].

In a related work, SpFN and RBD-ferritin nanoparticles (RFN) vaccination in mice and macaques were contrasted. After two doses in mice, RFN produced neutralizing titers that were more than 20 times greater than those in convalescent donor serum and were equal to those produced by a single vaccination with SpFN. Transgenic mice received strong protection from a deadly SARS-CoV-2 viral challenge after receiving a passive transfer of pure IgG from either SpFN- or RFN-vaccinated animals. Furthermore, immunizing rhesus macaques with two doses of RFN combined with a liposomal adjuvant resulted in nanoparticle albumin-bound titers that were 10 to 50 times higher than those shown in NHP studies with many licensed COVID-19 vaccines. Additionally, following a high-dose SARS-CoV-2 respiratory challenge, vaccination reduced virus replication in the upper and lower airways. The SpyTag/SpyCatcher system may also covalently bind SARS-CoV-2 protein antigens onto a protein nanoparticle core. *Streptococcus* pyogenes-derived SpyTag peptide (13 amino acids) and SpyCatcher proteins (116 amino acids) spontaneously form isopeptide bonds when combined[20]. SpyTag or SpyCatcher can be coupled to platforms for protein nanoparticles or vaccination antigens, enabling quick covalent bonding after mixing. SpyTag/SpyCatcher can improve high throughput testing of a variety of vaccination antigens or boost expression yields as compared to the direct fusing of antigens onto protein platforms. Using this method, ferritin nanoparticles (ferritin-NP-RBD) showing the SARS-CoV-2 RBD were created[21].

6.2.2. Liposomes: In contrast to lipid nanoparticles (LNPs), liposomes are nanostructured assemblies of amphipathic phospholipids that have an aqueous core inside one or more lipid bilayers that form a membrane. The antigens for the SARS-CoV-2 vaccination have been delivered through liposomes. RBD subunits were joined to the liposome surface to create RBD-liposomal vaccines in a preclinical study[22]. Histidine-tagged RBDs can create chelating interactions between cobalt ion and histidine residues by simply combining them with liposomes containing cobalt porphyrin-phospholipid (CoPoP) produced, causing RBD to exhibit on the liposome surface in a serum-stable, conformationally intact manner. In vaccinated mice, RBD-liposomes produced potent antibody titers

that prevented the reproduction of the live virus. Additionally, RBD-liposomes showed improved APC antigen absorption and improved immune cell recruitment to draining lymph nodes. Additionally, liposomes have been improved to improve their biomimetic capabilities. Compared to intramuscular or subcutaneous treatment, intranasal delivery of liposomes containing the Toll-like receptor agonist Poly (I: C) and coated with a pulmonary surfactant in mice generated a stronger mucosal immune response[23].

7. NANOPARTICLES VACCINE FOR COVID-19 IN CLINICAL TRIALS.

Although effective COVID-19 vaccines have been developed, the threat posed by SARS-CoV-2 and its variations still exists. The COVID-19 genome's coding sequences contain polymorphic alterations that have been discovered by scientists all across the world. The SARS-CoV-2 genome sequence alterations are known as VOCs because they have an impact on pathogenicity, vaccination sensitivity, and the efficiency of viral transmission. The development of effective SARS-CoV-2 vaccines has been significantly threatened and hampered by the emergence of VOCs. Up to this point, several VOCs that are referred to as Alpha, Beta, Gamma, Delta, and Omicron variations have been discovered and reported. Even though SARS-CoV-2 vaccines have had a lot of success, more work has to be spent on developing vaccines with a broad spectrum of effects to prevent infections brought on by new SARS-CoV-2 variants and weakened immunity[24].

A SARS-CoV-2 strain termed the Delta variant first surfaced in 2021 and quickly gained attention. The Delta variation has double the capability for transmission, a quick incubation period, and a higher viral load than the original strain. Therefore, there was a pressing need for the creation of vaccination against the SARS-CoV-2 Delta strain. Chen et al. demonstrated that RBD-conjugated NP vaccines produced a sufficient number of NAbs (neutralizing antibodies) as they protected mice (hACE2) against the Delta variant when administered using the single dose and prime-boost approaches. Additionally, the third booster dose of the trivalent vaccination in rhesus macaques led to the formation of wider cross-protective neutralizing antibodies. They concluded that RBD-conjugated NP vaccines are effective second-generation vaccinations against SARS-CoV-2 VOCs. A significant increase in NAbs against SARS-CoV-2 wild type and the Omicron variant was seen after the third dosage of the Walvax COVID-19 vaccine (ARCoV). An important reasonable strategy to combat the present rise of Omicron might be the administration of homologous booster vaccinations of ARCoV. The demand for next-generation vaccinations with broad-range protective effects against COVID-19 has increased globally as a result of the appearance of SARS-CoV-2 VOCs. Adjuvant SpFN (SARS-CoV-2 spike ferritin nanoparticle) vaccination has been produced and tested in non-human primates. Twice-administered SpFN (50 g) induced TH1 (T helper cell 1)-biased CD4 TH responses and elicited NAbs against wild type and VOCs[24].

Animal experiments have been conducted using the Omicron-specific LNP mRNA-based vaccination candidate alone and in combination with a heterologous booster of the wild-type mRNA vaccine. A new Omicron-specific LNP mRNA vaccine induced a substantial and targeted Ab response in mice that had never received vaccinations. Mice that had received a single booster dose of either a homologous booster injection of wild-type LNP mRNA or a heterologous booster shot of Omicron LNP mRNA after receiving a double dose of wild-type mRNA were able to restore their declining Abs responses to the Omicron variety. Results indicated a 40-fold increase in Abs after two weeks of booster dose treatment. Furthermore, compared to the homologous administration of wild-type booster against the Omicron variant, heterologous delivery of a booster dosage of LNP mRNA caused a 10- to 20-fold rise in Nabs[24].

8. PREVENTION OF CORONAVIRUS FROM ENTERING THE CELL

Due to the diversity in their genetic make-ups and the requirement for specialized antiviral medications, the creation of efficient and secure antiviral therapies for that particular strain of virus becomes increasingly difficult with the ongoing emergence of new and altered strands. In addition to helping to eliminate viruses already present in the body, the nanoparticles can also help to stop them from infecting cells. Nanomaterials like gold nanoparticles (gold NP) and carbon quantum dots (CQDs), which have a high specific surface area and may cling to a variety of antigens and/or substances on their surface, have been touted as potential instruments for interacting with viruses and inhibiting their entrance into cells[25].

A research work by Loczechin A. et al. showed how Boronic acid ligands conjugated with CQDs interfered with the coronavirus S protein's activity and significantly reduced the coronavirus' ability to infect host cells. It was noted that adding these NP to the cell culture media both before and after coronavirus infection significantly

decreased the rate of infection of the cells. Surprisingly, significant inhibitory activity was also found at the viral replication stage after one viral life cycle[26].

It was noted that adding these NP to the cell culture media both before and after coronavirus infection significantly decreased the rate of infection of the cells. Surprisingly, significant inhibitory activity was also found at the viral replication phase after one viral life cycle or 5.5 hours for coronavirus. Because they can easily enter the cell through endocytosis and interact with the virus's protein to stop viral genome replication, these nanomaterials with an average diameter of 10 nm and excellent water solubility emerged as promising contenders for defeating coronavirus[27].

9. DIFFERENT TYPES OF NANOPARTICLES TO COMBAT COV19/SARSCOV2

a. **Lipid Nanoparticles:** Four candidates for the LNPs encasing mRNA for the SARS-CoV-2 vaccine were created by BioNTech SE and Pfizer (BNT162a1, BNT162b1, BNT162b2, and BNT162c). They were created using two different kinds of nucleoside-modified mRNA, one that contained uridine and the other that self-amplified. They have undergone Phase 2 and Phase 3 clinical studies (NCT04380701 and NCT04368728, respectively) on healthy volunteers ranging in age from 18 to 85 years. The phase 3 clinical trials for the BNT162b2 candidate have concluded, and the vaccine appears to be both safe and efficacious. Pfizer and BioNTech filed their BNT162b2 to the FDA to ask for emergency use permission due to the urgent need for the Covid-19 vaccine. The Pfizer-BioNTech COVID-19 Vaccine was delivered in the United States on December 11, 2020, following FDA approval[28].

b. Gold nanoparticles: The diagnostic kit using gold nanoparticles makes it easy to identify an antigen from a swab. The conjugate pad contains the Au anti-SARS-2 antibodies. If SARS-2 antigens are present in the sample pad, they bind to the Au anti-SARS-2 antibodies and form antigen-antibody complexes during the run. One gold nanoparticle attaches to more than two anti-SARS-2 antibodies, which indirectly causes numerous antigens to bind to one gold nanoparticle, producing an intense color test line. This interaction is quite challenging. Accurate detection is aided by its vivid color. There aren't many Au anti-SARS-2 antibodies left in the conjugate pad. The SARS-2 antigens and Au anti-SARS-2 antibody complexes, along with free Au anti-SARS-2 antibodies, will rush to the test queue. Anti-SARS-2 antibodies at the test line are bound by Au anti-SARS-2, antibodies-SARS-2 antigens complexes, leaving unbound Au anti-SARS-2 antibodies. A new degree of complexity in Au anti-SARS-2 antibodies, SARS-2 antigens, and anti-SARS-2 antibodies leads to the creation of extraordinarily bright colors. The unbound Au anti-SARS-2 secondary antibodies in the control line simply bind to them and create color. These diagnostic kits are quite simple to recognize. The color of the test line indicates the presence of SARS-2 antigens. The color on the control line indicates the presence of Au anti-SARS-2 antibodies, demonstrating that the kit is working effectively. The control line's lack of color indicates that the kit is not in working condition. The test and control lines' colors indicate the presence of the antigen (a positive result), whereas the control line's color solely indicates the lack of the antigen (a negative result)[29].

c. Silver nanoparticles: AgNPs' precise mode of action for exerting their virus-killing impact is yet unclear. However, it has consistently been observed that AgNPs interact with the structural proteins on the surface of extracellular viruses to inhibit infection in the early stages, either by damaging the surface proteins to affect the structural integrity of virions, or by either entering viral attachment or preventing. Previous research demonstrated that AgNPs interfere with the viral entrance in the virus pre-treatment assay (VPrA), which successfully suppresses extracellular SARS-CoV-2 to shield the target cells from the infection and pseudovirus entry assay. By destroying the disulfide bonds on the angiotensin-converting enzyme-2 (ACE2) receptors and spike protein, AgNPs exert their antiviral impact on SARS-CoV-2. AgNPs can also have an intracellular antiviral effect by interacting with viral nucleic acids. Silver nanosponge's antiviral impact is size-dependent, with particles having a diameter of around 10 nm being the most potent. This suggests that interactions between 10 nm particles and viral proteins are more stable than interactions between bigger particles, which are not. The only AgNPs showing anti-SARS-CoV-2 action were those with sizes between 2 and 15 nm. This phenomenon of polyvinyl pyrrolidone-capped 10 nm silver nanoparticles (PVP-AgNP10) is supported by immunofluorescence research. Unlike AgNP100, it suppresses SARS-CoV-2.

Many Ag formulations may be consumed and inhaled that are being sold as a treatment for COVID-19. The over-the-counter treatment for COVID-19 is sold as a variety of inhalable and ingestible preparations of Ag. To

combat the COVID-19 pandemic, AgNPs can be applied to a variety of inanimate surfaces. Air filters from air conditioners and masks have been proven to be effective in suppressing SARS-CoV-2, suggesting that ag-coated devices might be used for medical equipment. It has been shown that polycotton textiles coupled with AgNP can prevent SARS-CoV-2[30].

d. Nitrous oxide(NO) nanoparticles: Viruses often have various sizes between 10 and 850 nm. Numerous bacteria, viruses, fungi, and tumor cells are known to be destroyed by NO. Applications for nitrous oxide and nanoparticles include a variety of fields. NO can be given in conjunction with other medications to effectively treat MERS-CoV-infected individuals and has shown promising results with minimal side effects. For COVID patients, NO inhalation therapy has been studied in the United States of America and China. In the past, NO was successfully utilized to treat lung-related illnesses brought on by certain viruses. According to the investigations, NO compounds may be able to stop SARS-CoV replication. S-nitrosoN-acetyl penicillamine, a nitrous oxide donor, effectively inhibited the SRS-CoV replication pathway. Many researchers attempted to evaluate the effects of NO against SARS-CoV-2, and NO donors were viewed as crucial medical interventions[31].

10. FUTURE PROSPECT

To battle the COVID-19 pandemic, RNA and LNP-based vaccines were quickly developed and received emergency FDA approval, demonstrating the quick and efficient responsiveness of this strategy against complicated illnesses. These vaccines have been created for a variety of disorders, including cancer and hyperlipidemia, in addition to infectious infections. Several clinical trials are nearly finished. There is already much research underway, and there will be more in the future, which portends the release of a variety of goods. Additionally, mRNA and protein Because NPs vaccines need sophisticated production facilities, they are a concern in low-income nations. Protein NPs have unique difficulties, such as difficult purification, stability problems, and strict GMP-grade cell line requirements, which make getting regulatory clearances difficult. Technology transfer is necessary to scale up the manufacturing of the NPs vaccine and its related advantages to guarantee a plentiful supply of produced vaccinations across the world. For the treatment and prevention of COVID-19, NPs vaccines enable regulated antigen release and viral entrance inference. Clinical results may be used to assess the NPs vaccine's efficacy, stability, and safety. Conducting ongoing experiments to confirm the security and efficacy of NPs vaccinations is necessary.

Due to extensive research into the development of various vaccine platforms, it was possible to start NP-based clinical studies within two months of the public release of the SARS-CoV-2 genome sequence. In actuality, the rapid development of SARS-CoV-2 vaccinations and their efficiency contributed to saving many lives. However, only time will tell how these NP-based vaccines develop in the upcoming COVID-19 phase and/or in the face of pandemic situations.

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