Microneedles for SARS-CoV-2 Mass Vaccination

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Vaccination is an effective measure to prevent infectious diseases. Protective immunity is induced when the immune system is exposed to a vaccine formulation with appropriate immunogenicity. However, traditional injection vaccination is always accompanied by fear and severe pain. As an emerging vaccine delivery tool, microneedles overcome the problems associated with routine needle vaccination, which can effectively deliver vaccines rich in antigen-presenting cells (APCs) to the epidermis and dermis painlessly, inducing a strong immune response. In addition, microneedles have the advantages of avoiding cold chain storage and have the flexibility of self-operation, which can solve the logistics and delivery obstacles of vaccines, covering the vaccination of the special population more easily and conveniently.

Keywords: immune response ; microneedles ; mass vaccination ; SARS-CoV-2 vaccine

1. Development and Selection of (Microneedles) MNs

1.1. Classification of MNs

MNs generally fall into five different depending on their delivery strategies: solid, coated, hollow, dissolving, and hydrogel-forming.

Solid MNs were the first generation and were made of metal materials and non-degradable polymers such as silicon and titanium dioxide. They are generally described as “poke and patch” due to their inability to carry the drug [1]. They can pierce the skin, leaving a microchannel that allows drugs or vaccines to reach the dermis. Coated MNs were proposed to overcome the complexity of the two-step process in solid MNs [2]. They load the active substances through the coating on the surface of the needle body, then release the deliverables into the capillaries through the intercellular fluid, and finally induce systemic treatment. They are described as “Coat and poke”, and the substances delivered are mainly water-soluble, which can be reused [3]. An example is the successful preparation of smallpox vaccine-coated MNs by Chou et al., which turned out to be an alternative delivery system for traditional smallpox vaccination and storage [4]. The hollow MNs can be understood as a microsyringe due to the cavity in the middle of the needles. When they pierce the skin, the vaccines preloaded in the cavity are driven by the concentration gradient to achieve delivery. They are usually made of hard materials such as silicon, metals and polymers [5].

All three types of MNs mentioned above are at risk of breakage and generate sharps waste, which prompted the development of dissolving MNs [6]. They are made of water-soluble biocompatible/biodegradable polymers or sugars. Dissolving MNs are often described as “Poke and release”. After penetrating the skin, the needles gradually dissolve in the skin tissue, and the substances contained in the needles are gradually released and absorbed by the body. Because of their advantages in delivering various therapeutic agents and vaccines, dissolving MNs have become widely used throughout the MN industry. Hydrogel MNs are usually made of cross-linked hydrogels or super-swollen polymers [7]. They are used to deliver drugs by swelling or to diagnose by absorbing certain substances from interstitial fluid in the skin without dissolving.

1.2. Materials for the Preparation of MNs

The main attribute of MNs is their ability to penetrate the skin without breaking or bending. At present, the materials used for MNs are divided into three categories: inorganic materials, metals, and polymers.

Inorganic materials used in MNs include mainly silicon, glass, and ceramics. Silicon is the first and most frequently used material, which can be customized in different shapes and sizes for a wide range of applications. Although it has high hardness and can be easily inserted into the skin, silicon is brittle and can cause adverse effects if left in the skin [8]. Because of this, scientists have used glass as a material to make MNs. Glass is now widely used for alumina, zirconia and calcium sulfate hemihydrate [9][10][11]. However, glass is brittle and fragile. Scientists have begun to investigate

biocompatible ceramic materials. For example, Vallhov et al. evaluated biodegradable ceramic (calcium sulfate) MNs and found that they could release drugs without triggering an immune response [12].

Metals have been used in medical devices for decades. Biomedical metallic materials have good biocompatibility; gold, silver, platinum, stainless steel, cobalt-based and titanium-based alloys, etc. have mechanical properties useful in dentistry. Nickel-chromium stainless steel, cobalt-chromium-molybdenum alloys, titanium and their alloys are used in orthopedics, which are suitable materials for MNs. Among these, stainless steel, titanium, and nickel are often used for metal MNs [13][14][15]. However, due to high hardness, non-biodegradability and inorganic materials, metals can only be used for solid, coated, or hollow MNs. Scientists further developed high molecular polymers to make dissolving and hydrogel MNs.

Polymer is a very promising material for MNs. Various biocompatible and biodegradable polymers have also been used to prepare MNs, such as hydrophobic poly (methyl methacrylate) (PMMA), Polyvinyl alcohol (PVA), Polyvinylpyrrolidone (PVP), polycaprolactone (PCL) and hydrophilic hyaluronic acid (HA), Carboxymethyl Cellulose (CMC), Polyethylene glycol (PEG) et al. [16]. Polymers are not as rigid as silicon and metals, and they are commonly used to make dissolving and hydrogel MNs [17].

2. Development of MNs Delivery of Vaccines

2.1. Classification of Vaccines

Vaccines approved for human use in the pharmaceutical industry today fall into four main subcategories: live-attenuated vaccines, inactivated vaccines, pathogen component vaccines (DNA, RNA, proteins, virus-like particles (VLPs), etc.), and toxoid vaccines. Among them, live attenuated and inactivated vaccines are whole-agent vaccines, and pathogen component vaccines are subunit vaccines.

2.2. MNs Deliver Different Types of Vaccines

Mikszta et al. [18] first tried MN immunization with a novel MEA in 2002 and successfully broke the skin barrier to deliver the vaccine to the body. Research on the delivery of vaccines via MNs has been intensively pursued after that.

2.2.1. Live-Attenuated Vaccine

Live-attenuated vaccines are whole vaccines in the form of weakened “wild” viruses or bacteria. When introduced into the body, they can trigger an immune response as with normal bacteria or viruses without causing harm or disease. However, these weakened forms are very likely to mutate back to the former wild type and cause fatal damage to the organism. Oral poliovirus vaccines, for example, can revert to their original toxic form, which can enter the central nervous system and cause paralysis in patients [19]. In addition, vaccines are highly unstable at temperatures above 8 °C. Professional operations are also required to maintain the cold chain for vaccine storage and transport. The use of MNs can reduce the need for low temperatures while ensuring a more stable state of vaccines, and it is safer and more convenient to operate.

In a study using the attenuated Japanese encephalitis vaccine ChimeriVax in non-human primates, it was found that the same dose of vaccine administered via skin micro abrasion and penetration with MNs produced stronger immunity than from subcutaneous injection [20]. In addition, Edens et al. used dissolving MNs to deliver a live-attenuated measles vaccine to rhesus monkeys and found that MN vaccination produced higher antibody titers than subcutaneous vaccination. In addition, compared with commercially available freeze-dried and liquid vaccines, which can be stored for 90 days and only 7 days, respectively, the MN measles vaccine can retain about 90% of its potency after being stored at 40 °C for 4 months [21]. Prausnitz et al. also summarized the benefits of using MNs for measles and rubella vaccination, including the simplification of supply and cold chain, the elimination of needlestick waste, and the reduction of vaccination system costs. Hiraishi et al. used a microneedle patch (MNP) to efficiently deliver a BCG vaccine into the epidermis and dermis of the skin and elicit a robust cell-mediated immune response in the lungs and spleen of guinea pigs. This approach not only simplifies logistics and eliminates the hazards posed by hypodermic needles, but also promises to increase BCG vaccination rates [22].

2.2.2. Inactive Vaccine

Inactivated vaccines are produced by inactivating intact viruses or bacteria by chemical methods (beta-propiolactone, formalin, etc.) or physical methods (ultraviolet rays, electron beam irradiation, etc.) [23]. Although this kind of vaccine is not effective as the live-attenuated vaccine, it is safer. Inactivated vaccines can induce massive immune responses by

multiple injections or co-injection with adjuvants. Hepatitis A, hepatitis B, and influenza vaccines are all inactivated vaccines [24]. To date, there are many examples of the successful use of MNs to deliver inactivated vaccines.

Hirschberg et al. used dissolving MNs to inject a hepatitis B vaccine containing aluminum hydroxide and lipopolysaccharide (LPS) adjuvant, and its primary immunization effect was comparable to the secondary immunization effect of a conventional vaccine containing liquid alum adjuvant [25]. Moreover, the conventional vaccine lost 40% of its antigenicity after one week at 50 °C, whereas the MN vaccine did not significantly decrease after three weeks at 50 °C. This example successfully demonstrates that MN may be the most promising alternative to needle injection for hepatitis B vaccines. Frewab et al. evaluated the acceptability of an inactivated influenza vaccine delivered by MNP compared with an inactivated influenza vaccine (IIV) delivered by hypodermic needle [26]. They screened 112 normal people in Atlanta, Georgia, and they found that participants also preferred the MNP for influenza vaccination and follow-up immunizations. Rodgers et al. successfully inserted and dissolved MNs ranging in size from 254 to 381 microns into the skin of mice. The results demonstrated that the bacterial load in the lungs of mice that had been inoculated with Pseudomonas aeruginosa was significantly lower than that of uninoculated mice.

2.2.3. Pathogen Component Vaccine

Pathogen vaccines are subunit vaccines and contain antigenic parts of viruses and bacteria. It can trigger an immune response without harming the subject [27]. Pathogen vaccines include DNA, RNA, protein, and VLPs, and they resemble inactivated vaccines but are less immunogenic than live viruses or bacteria. They are safer but require multiple doses to achieve the desired level of the immune response [28].

DNA Vaccine

DNA vaccines have been studied since the early 1990s. They are antigen-encoding plasmid vectors containing a gene of interest [29]. When the plasmid is transfected into myocytes or inoculated into APCs in skin or muscle, it triggers the transcription of the gene and the production of an antigenic protein, eliciting antigen-specific immune responses in vivo [30]. The advantages of DNA vaccines are that they are more heat-resistant than traditional forms of vaccines and can be easily mass-produced [31]. DNA vaccines cannot be reverted to their original virulent form [32].

Arya et al. evaluated the safety and immunogenicity of MNP vaccination with a rabies DNA vaccine [33]. The vaccine in the MNP was stable at 4 °C for at least three weeks, and the MNP was well tolerated in the skin, with complete resolution of skin reactions within seven days and no systemic side effects. The immunogenicity of the MNP outperformed the intramuscular injection of the same vaccine dose. Dissolving MNPs may provide an innovative approach for mass rabies vaccination [34]. Cole et al. used MNs to enhance the immunogenicity of DNA vaccines [35]. This approach not only improves DNA stability in solid matrices, but also increases DNA delivery ability compared to sMN. To achieve an effective transdermal vaccine and targeted delivery in developing countries, Hu et al. utilized the MN delivery of a DNA vaccine for the treatment of malignant melanoma. The results showed that the MN-delivered vaccine induced significant therapeutic anti-tumor immunity and inhibited cancer cells growth, which is a potential immunotherapy strategy for MM [36]. Liao et al. synthesized DNA multiplex vaccines in a single step in an MNP. It can be stored at 45 °C for at least 4 months, which has a significant impact on effective vaccination in developing countries [37]. In addition, Qiu et al. developed a hepatitis B DNA vaccine system based on a soluble microneedle array that can induce an effective immune response [38].

RNA Vaccine

RNA vaccines function by introducing an mRNA sequence that encodes a specific antigen [39][40]. The vaccine introduces molecules of antigen-encoding mRNA, and the designed mRNA serves as a blueprint for building a foreign protein that would normally be produced by a pathogen (e.g., a virus) or by a cancer cell. These protein molecules stimulate an adaptive immune response that teaches the body to identify and destroy the corresponding pathogen or cancer cells [19]. The mRNA is delivered by a co-formulation of the RNA encapsulated in lipid nanoparticles that protect the RNA strands and facilitate their uptake into cells [41].

Since Nair and Boczkwoski first successfully demonstrated mRNA-based cancer vaccination in 1996 [42], scientists have been searching for the best mRNA delivery system in recent years. MNs have attracted considerable interest as a platform for delivering vaccines through the skin. Koh et al. reported a proof-of-concept study to produce, characterize, and therapeutically evaluate in vitro transcribed messenger RNA (mRNA) loaded into dissolving MNP (RNA patches) [43]. They found that the physical and functional integrity of mRNA stored in the MNs were preserved for at least two weeks. The RNA MNP can mediate in vivo transgene expression of mRNA encoding luciferase for up to 72 h, and the transfection efficiency and kinetics are superior to subcutaneous injection. Golombek et al. evaluated the intradermal delivery of synthetic mRNA by injection with hollow MNs. In addition, an in vitro porcine skin model was established to analyze

protein expression mediated by synthetic mRNA in the skin after intradermal administration [44]. Using this model, the efficient delivery of synthetic mRNA was demonstrated to detect high levels of secreted humanized Gaussian hypolucidase (hGLuc) protein encoded by microinjection of synthetic mRNA. The use of MNs enables the patient-friendly, painless, and efficient delivery of synthetic mRNA into the dermis. This approach can be used for topical treatment of different skin diseases as well as for vaccination and immunotherapy. There are only two typical examples of RNA vaccine delivery using MNs so far.

Protein Vaccine

Protein subunit vaccines contain specific isolated proteins from viral or bacterial pathogens to trigger protective immunity [45]. Rather than injecting a whole pathogen to trigger an immune response, a protein vaccine is safer and more stable but more complex to manufacture. Generally, adjuvants are required to induce a strong immune response, and multiple injections are performed.

MNs can be used as a delivery tool to deliver protein vaccines efficiently. Yuan et al. delivered the F1 protein antigen of Yersinia pestis in MNs, which were successful in triggering an immune response against the plague in animals [46]. Weldon et al. tested the hypothesis that a recombinant subunit influenza vaccine could be delivered to the skin via coated MNs [47]. It was found that mice vaccinated with stable recombinant trimeric soluble hemagglutinin (sHA) by MN elicited a strong immune response. The mice were completely protected from lethal influenza virus infection, highlighting the benefits of this protein subunit vaccination. Wang et al. generated self-adjuvant protein nanoparticles that conserved influenza antigens and immunized mice by vaccinating the skin with dissolvable MNPs to enhance the strength and breadth of the immune response [48]. They produced a bilayer protein nanoparticle, NA2-FliC/M2e, and demonstrated that this nanoparticle-based MNP skin vaccine could be developed as an independent or synergistic component of a universal influenza vaccination strategy.

VLP Vaccine

VLPs are nanoparticles composed of a subset of non-infectious viral components that are structurally similar to wild-type viruses but lack the viral genome. They are non-replicative and non-infectious, and can induce an immune response in the host. VLPs have good stability and are excellent vaccine carriers [49][50].

Quan et al. used coated MNs to deliver influenza VLPs into mouse skin [51]. It was found that the delivery of high doses of vaccine via MNs resulted in 100% protection against challenging influenza viruses. In contrast, unstable influenza VLPs and intramuscular vaccines weakened the immune system and provided only partial protection (≤40%). A vaccine formulated with a coated MNP was shown to provide superior protection over intramuscular injection through dermal vaccination with potential dose maintenance. Ray et al. developed dissolving MNs that contained a candidate HPV vaccine consisting of Qβ VLPs [52]. Compared with conventional subcutaneous injection, polymer MN delivery of Qβ-HPV produced similar levers of anti-HPV16 L2 IgG antibodies, with a lower (16.7%) intradermal dose required. In addition, the vaccine can remain stable in the MNs at room temperature for several months, which will effectively solve many problems related to the cold chain. This MNP vaccine not only saves vaccine doses, but is also easy to self-administer and minimally invasive, which enables the wide distribution of HPV vaccines and improves patient compliance. The Qβ-VLPs and their MN delivery technology are a plug-and-play system that can serve as a general platform with a wide range of applications. Guo et al. developed a novel tumor vaccine delivery strategy using a biodegradable microneedle patch (MN) that allows for the sustained release of tumor antigens and induces long-term antitumor responses [53]. Kines et al. also demonstrated that mice immunized with HPV16 VLP-coated microneedles produced a robust neutralizing antibody response, and the MN delivery of freeze-dried HPV may provide a practical, heatable vaccine delivery method that can be evaluated clinically [54].

2.2.4. Toxoid Vaccines

Toxoid vaccines use toxins (harmful products) produced by disease-causing bacteria that develop immunity against the disease-causing part of the bacteria rather than the bacteria themselves. A toxoid is an inactive toxin that has lost its ability to cause disease but retains its immunogenicity [55]. Two vaccines contain toxoids as immunogens, namely the diphtheria and tetanus vaccines. Like some other vaccines, the toxoid vaccines require booster shots to provide lasting protection against disease. In recent years, numerous studies have been conducted on the use of microneedles to deliver toxoid vaccines [56].

Groot et al. tested ceramic nanoporous microneedle arrays (MNAs), which are a novel MN drug delivery technology capable of delivering diphtheria toxoid (DT) and tetanus toxoid (TT) in vivo [57]. The results showed that DT and TT can be successfully loaded into the tips of ceramic nanoporous MNs. By labeling the antigens with near-infrared fluorescence,

they applied DT and TT-loaded nanoporous MNAs to mice skin in vivo and induced antigen-specific antibodies similar to those obtained with subcutaneous immunization at the same doses, opening the possibilities for future MN vaccination designs. Leone et al. achieved single and multiple injections of DT using dMNs and hMNs [58]. The prepared dMN can penetrate the skin and be dissolved within 20 min to release the antigen all at once. Skin immunization with hMN was performed by repeated dose injections. The overall response to dissolved MN vaccination was higher than that of hMN, and the immune strength was also high in the absence of adjuvant. In conclusion, unadjuvanted dissolving MNs were proved to be promising delivery vehicles for vaccination. Pattarabhiran et al. investigated potent immune responses elicited by tetanus toxoid-antigen-dissolvable microneedles (TT-MN) in a mouse model [59]. They prepared TT-MN by adding TT to the polymer mixture. The results showed that the MNs penetrated 130 microns deep into the mouse skin and dissolved completely within 1 h of insertion into the skin. The TT-MN group had higher antibody titers than the intramuscular injection group. This indicates that TT-MN can be developed as a minimally invasive system for percutaneous delivery of TT antigen.

3. COVID-19 Vaccines and Their MNs Delivery

3.1. SARS-CoV-2

SARS-CoV-2 is a coronavirus strain of the respiratory disease that caused the COVID-19 pandemic [60]. It is a positive single-stranded RNA virus with an envelope [61] and belongs to the family Coronaviridae Betacoronavirus genus severe acute respiratory syndrome related coronavirus species [62]. SARS-CoV-2 is the seventh-known coronavirus that can infect humans. Available evidence suggests that it is a zoonotic coronavirus, with close genetic similarity to bat coronaviruses [63]. Research is ongoing as to whether it came directly from bats or indirectly from any intermediate host [64]. The virus showed little genetic diversity, suggesting that a spillover event introducing SARS-CoV-2 into humans may have occurred in late 2019 [65]. Epidemiological studies estimated that the primary reproduction number (R0) of SARS-CoV-2 was on average 2.4 to 3.4 between December 2019 and September 2020 [66]. However, some subsequent variants became more contagious, such as Delta (B.1.617.2 and AY lineage) and Omicron (B.1.1.529, BA.1, BA.1.1, BA.2, BA.3, BA.4 and BA.5 lineages).

The virus can invade the human body through the upper respiratory tract, and it uses the Angiotensin-converting enzyme 2 (ACE2) receptor to enter host cells. The main infected organs include the lungs, heart, kidneys and other major organs [67]. Human patients infected with the virus have no unique clinical symptoms, most of which are low-grade fever, weakness, oral and nasal symptoms, and dry cough, and some are accompanied by gastrointestinal discomfort [68].

3.2. Types of COVID-19 Vaccines

In the aftermath of the outbreak of the pandemic, various scientific research units and vaccine companies have been working on developing various COVID-19 vaccines, all of which can teach the human immune system to safely recognize and block the virus. As of Oct 2022, there are more than 700 vaccines under development in preclinical or clinical trials, and more than 220 COVID-19 vaccine candidates are in development. Among them, at least 85 vaccine candidates are in human trials, and 40 vaccines have been approved by the FDA for production use.

There are two broad categories of COVID-19 vaccines, including whole viral vaccines and component viral vaccines similar to those described before. Whole-virus vaccines include inactivated vaccines and live-attenuated vaccines; component vaccines include recombinant subunit vaccines, viral vector vaccines, and nucleic acid vaccines.

3.3. MN Delivery of COVID-19 Vaccine

In 2020, Kim successfully delivered the COVID-19 recombinant protein vaccine using dissolving MNs, which kicked off this field [69]. As of October 2022, a total of 44 related papers have been published.

3.3.1. Whole-Virus Vaccine

Whole-virus vaccines are made by using chemical or physical methods such as formaldehyde or heat to kill the viral pathogen completely or partially, followed by purification and the addition of adjuvants. Such systems are very mature and are a massive production. It is a priority development technology for vaccines in response to COVID-19 outbreaks. Many whole-virus vaccines are currently available, such as BBIBP-CorV developed by the Beijing Institute of Biological Products (BBPI) of China Pharmaceutical Group [70]. CoronaVac (also known as the Sinovac COVID-19 Vaccine) was developed by Sinovac, a biopharmaceutical company in mainland China [71][72]. The Indian COVID-19 vaccine Covaxin (BBV152) was created by Bharat Biotechnology and the Indian Council of Medical Research [45]. The WHO included the above three vaccines on the “Emergency use list” in May, June, and November 2021, respectively.

Li et al. developed a smart mushroom-inspired printable and mildly detachable (MILD) MN platform for the efficient and convenient delivery of multiple-dose COVID-19 vaccines and decentralized vaccine information storage. The MILD system induced high levels of antibodies in vivo after loading with an inactivated SARS-CoV-2 virus vaccine, which is a promising vehicle that has the potential to help contain the COVID-19 pandemic [73].

3.3.2. Recombinant Subunit Vaccine

Recombinant subunit vaccines contain only selected parts of pathogens and are very safe and stable. The expression vector containing the target antigen gene is transfected into an engineered cell line, and large quantities of the expression-target protein is purified; finally, the recombinant subunit vaccine can be prepared after adding an adjuvant. The preparation technology of recombinant subunit vaccines is mature, and there are currently more than 50 protein subunit vaccines in development [74]. In clinical trials, the overall protection rate of subunit vaccines is higher than that of inactivated vaccines, which are also suitable for immunocompromised individuals. However, their manufacturing is more complex and requires adjuvant co-injection.

The first vaccine to be successfully delivered using MNs was a recombinant protein vaccine [69]. Afterwards, Kuwentrai et al. prepared MNs with low molecular weight hyaluronic acid (HA) as support material for the delivery of S-RBD protein vaccines. HA is a naturally occurring skin substance that can be rapidly dissolved in skin tissue fluid. The results showed that the MN-based minimally invasive intradermal vaccine effectively penetrated the skin of mice, eliciting significant B-cell antibody responses and inducing interferon-gamma (IFN-γ)-based T-cell responses, which may control the rapid COVID-19 outbreaks [75]. McMillan et al. used a high-density microarray patch (HD-MAP) to deliver a SARS-CoV-2 spike subunit vaccine directly to the skin, which indicated that the vaccine was thermostable on the patch and enhanced cellular and antibody immune responses. In an ACE2 transgenic mouse model, a single dose of HD-MAP-delivered spikes provided complete protection against a lethal viral challenge. HD-MAP-delivered vaccines are superior to traditional needle and syringe vaccinations and could be an important addition to the ongoing COVID-19 pandemic [33].

3.3.3. Viral Vector Vaccines

Viral vector vaccines are produced by constructing a viral vector containing the target antigen gene and then delivering the genetic material that encodes another infectious pathogen-targeting antigen to the recipient's host cell. It provides the genetic material to express antigens in cells and can induce a powerful cytotoxic T cell response. Adenoviral vectors are the most used viral vectors for this vaccine.

This type of vaccine mainly includes AstraZeneca, Satellite V, Johnson & Johnson, Convidecia, etc. Oxford–AstraZeneca (AZD1222) is an improved non-replicating chimpanzee adenovirus vector (ChAdOx1) vaccine developed by the University of Oxford in cooperation with AstraZeneca Pharmaceuticals [76]. This vaccine was placed on the emergency use list by the WHO on 15 February 2021 [77]. Satellite V (Спутник V) is a vaccine developed and registered by the State Research Center of Epidemiology and Microbiology in Gamaleya, Russia. Johnson & Johnson's COVID-19 vaccine is a human adenovirus-based viral vector vaccine developed by Janssen Vaccines in Leiden, the Netherlands and Janssen Pharmaceutica. Ad5-nCoV (Convidecia) is a single-dose recombinant adenovirus type 5 vector vaccine developed by CanSino Biological Co., Ltd. and the Institute of Bioengineering of the Academy of Military Medical Sciences of the Chinese People's Liberation Army Academy of Sciences [78]. Ad5-nCoV is the only currently approved COVID-19 vaccine that can use a single-dose vaccination program and is approved in China [79]. Flynn et al. identified a dissolvable MNP for skin immunization to deliver the malaria vaccine AdHu5-PfRH5. Studies have shown that MNs can deliver low-dose adenovirus vaccines that are highly immunogenic. Moreover, the MNs also can stabilize the adenovirus vaccine [80].

3.3.4. Nucleic Acid Vaccines

Nucleic acid vaccines introduce specific antigen-encoding DNA or mRNA sequences into the cells of an organism to induce an immune response, preventing and treating diseases. Compared with traditional vaccines, genetic vaccines have the advantages of convenient design, high speed, and low production cost. Currently, there are more than 30 types of COVID-19 mRNA vaccines in preclinical or clinical trials around the world.

The Pfizer–BioNTech vaccine (BNT162b2, trade names: Comirnaty, Fubitai) is an mRNA vaccine jointly developed by BioNTech in Germany and Pfizer in the United States. It is the first nucleic acid vaccine approved by the WHO for the prevention of COVID-19 [81]. The Moderna vaccine (mRNA-1273, trade name: Spikevax) was jointly developed by the National Institute of Allergy and Infectious Diseases at the Biomedical Advanced Research and Development Authority and Moderna Corporation [82]. On 18 June 2022, the U.S. Food and Drug Administration (FDA) issued an emergency authorization for the use of Moderna and Pfizer–BioNTech vaccines in infants and young children over 6 months of age

[83]. The above two vaccines have launched mass vaccinations around the world; both of them have proven to be highly effective.

3.4. Advantages and Challenges of MN Delivery of COVID-19 Vaccine

The development of MN undoubtedly provides a convenient alternative vaccination method for addressing the COVID-19 pandemic, which is conducive to increasing the popularization rate of the vaccine, especially at this stage when COVID-19 vaccination has been vigorously strengthened. There are many advantages to using MNs to inoculate the COVID-19 vaccine. It could not only lower the inoculation dose of the vaccination, but it also induces higher antibody levels. Because it is less invasive, easy to operate by itself, and has enhanced thermal stability, it has the potential to expand vaccination rates in low- and middle-income countries and special populations (infants, the elderly, and pillow phobias, etc.), which are expected to achieve full vaccine coverage.

However, there are currently many challenges for the MN delivery of the COVID-19 vaccine. During the drying, curing, and loading process of the MNs, the loading of the vaccine is difficult to make uniform, and there is great instability. Therefore, the corresponding antibody titers are also unstable. For sMNs or hMNs, potential safety hazards may be caused by needle tip breakage. Whether in the process of manufacturing or in storage, sterility is difficult to guarantee. Most importantly, there is little clinical data to support the immune response after vaccination, and there is long way to go before practical application.

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