International Journal of Dermatology

Updates from medicine

# COVID-19 vaccines and nanomedicine

**Robert S. Shapiro, MD**

Private Practice, Hilo, HI, USA

**Correspondence**
Robert S. Shapiro, MD
3242 San Amadeo, 2 F
Laguna Woods, California 92637
E-mail: robertsethshapir@icloud.com

Conflict of interest: None.

Funding source: None.

doi: 10.1111/ijd.15673

## Abstract

**Background** The COVID-19 virus-induced pandemic has been the deadliest pandemic to have occurred in two generations, besides HIV/AIDS. Epidemiologists predicted that the SARS-Cov 2 pandemic would not be able to be brought under control until a majority of the world's population had been inoculated with safe and effective vaccines. A world-wide effort to expedite vaccine development was successful. Previous research for vaccines to prevent SARS and MERS, also coronaviruses, was vital to this success. Nanotechnology was essential to this vaccine development. Key elements are presented here to better understand the relationship between nanomedicine and the COVID-19 vaccine development.

**Methods** NLM PubMed searches for COVID-19 vaccines, nanotechnology and nanomedicine were done. There were 6911 articles screened, 235 of which were deemed appropriate to this subject and utilized here, together with two landmark nanomedicine texts used to expand understanding of the basic science of nanotechnology.

**Results** SARS-Cov 2, caused by the COVID-19 virus, was first recognized in China in December of 2019 and was declared as a pandemic in March of 2020. The RNA sequence was identified in January of 2020. Within 4 months of the viral genome being released, over 259 vaccines had been in development. The World Health Organization (WHO) anticipated a vaccine with a 50-80% efficacy to be developed within 1-2 years. Ahead of schedule, the Food and Drug Administration (FDA) announced the emergency authorization approval for two mRNA vaccines within 11 month's time. Nanotechnology was the key to the success of these rapidly developed, safe and effective vaccines. A brief review of pertinent basic science principles of nanomedicine are presented. The development of COVID vaccines is reviewed. Future considerations are discussed.

**Conclusions** Control of the COVID-19 SARS-Cov2 pandemic benefitted from nanomedicine principles used to develop highly effective, yet very safe and relatively inexpensive vaccines. These nanovaccines can be much more easily altered to adjust for viral variants than traditional live or inactivated legacy-type whole virus vaccines.

The U.S. government promoted SARS-CoV-2 vaccine development. The World Health Organization (WHO) anticipated a vaccine with a 50–80% efficacy.[1] The vaccines were optimistically predicted to be available in 1–2 years. Vaccine development began as soon as the RNA sequence for the SARS-CoV-2 virus was identified. Ahead of schedule, the Food and Drug Administration (FDA) announced an emergency authorization approval for two nanovaccines 11 months later, which have about a 95% efficacy. This fortunate success was built on previous developments in nanomedicine. The initial timeframe estimates were based on previous experience with live, attenuated, or killed vaccine, all legacy technologies. The nanoparticle carriers are integral to the success of the mRNA vaccines. In addition, mRNA itself can act as an adjuvant.[2] The necessity to develop a safe, rapidly available, scalable, inexpensive, and effective vaccine to control the COVID-19 pandemic has thrust nanotechnology into the forefront, displaying its potential to save millions of lives. RNA therapies first started in 1998.[3] The National Nanotechnology Initiative (NNI) was initiated back in 2000.[4] A thorough review of nanovaccines was published in 2009.[5]

Over 136 million cases have been documented, and 2.94 million people have died worldwide due to the COVID-19 pandemic as of April 12, 2021. COVID-19 is currently a leading cause of death in the United States.[6] Two earlier 21st century betacoronavirus epidemics, MERS and SARS, were more easily brought under control due to the fact that only symptomatic patients were contagious with those infections. COVID-19 became a pandemic due to the inability and impracticality of frequently testing the whole world's population, as the pandemic was fueled by travel and asymptomatic transmission.

It is commonly quipped that if we can send a man to the moon (12 men so far) and fly to Mars (26 successful flights),

why can we not cure the common cold, 20% of which are due to a coronavirus?[7,8] The coronavirus was first discovered in the same decade as men walked on the moon.[7] This was in a time before fax machines or microwave ovens, in the days of giant room-sized mainframe computers that were capable of an infinitesimally small functional capability compared to a current smart phone. The nascent space technology was sufficient for moon walking in the 1960s; however, until the human genome project and the development of biotechnology and nanotechnology, there was no remote possibility of attempting to cure the common cold.

Segueing into the 21st century, the cold war has fortunately been transformed into a more societally beneficial and peaceful "cold" race for a COVID-19 vaccine. The Russian COVID-19 vaccine, named SPUTNIK V, has arrived[9] (without completing the third phase of more widespread testing[10]) several months in advance of the first two U.S. mRNA nanovaccines.

One of the most important issues regarding the severity of the COVID-19 pandemic relates to the high Ro,[1,11] which is due to the high degree of contagion of asymptomatic patients. The immunopathology of COVID-19 is not fully understood. The virus has many mechanisms of immune evasion.[10,12] Critically, there is asynchrony[13] and dysfunction[8,14] of the patient's immune system, leading to hypercytokinemia,[15] enormous amounts of IL-6[8,16,17] with contributions by IL-17 from Th17 T cells[16,18] and TNF alpha,[17] as well as effects on PD-L1 and CTLA-4. Endoplasmic reticulum stress, T-cell exhaustion,[16] macrophage activation syndrome, acute respiratory distress syndrome (ARDS), thrombotic complications due to complement triggered microangiopathy, and disseminated intravascular coagulation (DIC) with elevations of D-dimer[19] can occur. Death is often due to this inappropriately exuberant immune response.

## Nanotechnology

Nanotechnology is a form of biomimetic technology. Viruses are themselves nanoparticle sized. Therefore, basic understanding of nanotechnology is essential. There are 1927 articles in a PubMed search related to COVID-19 and nanotechnology as of April 12, 2021. Nanomedicine exists at the intersection of bioengineering, pharmacology, quantum mechanics, biology, nuclear physics, photonics, materials science, computational science, and radiation.[20,21]

Nano is defined as size equal to or less than 100 nm. Visible light has a wavelength of 400–700 nm. Nanoparticles approach molecular sizes (1000 nm = 1 micron) (500 Daltons = ~2.5 nm).

Particles that are close to molecular size do not act in the same way as their micro or macro forms. It is analagous the phase differences of solid, liquid, gas, plasma, as well as the differences between astronomic bodies and subatomic particles. For instance, carbon[22] and salt are electrical conductors as nanoparticles. Gold is a liquid as a nanoparticle.[23] Glass is flexible in nanoparticle sizes. The interparticle interactions, surface attractions, ionization potential, surface reactivity, and magnetism all change when materials approach molecular dimensions. Fifteen and 50 nm gold nanoparticles can easily penetrate the blood brain barrier[24] and be detected 6 months after IV injection. Nanoparticle penicillin is effective against MRSA.[25]

Nanomaterials exist naturally in the environment, and they can be manufactured. Vog, air pollution, auto emissions, and cigarette smoke are composed of nanoparticles. Soot and diesel smoke are primarily comprised of carbon nanoparticles. Nanoparticles are already in widespread use in industry. There are at least 424 reported uses of nanotechnology in cosmetics and personal care products. There are 250 clothing companies that offer clothing with nanotechnology. Nanosilver is in socks, and nanoparticles are in food storage containers, washing machines, soaps, surgical masks, food, paint, coatings, and tires. Carbon nanotubes are in tennis rackets and bicycle frames. Hong Kong sprayed its whole subway system's areas of contact with a nanoparticle antibacterial agent. Quantum dots are in solar cells.

Military textiles are being created to detect chemical and biological weapons, perform physiologic monitoring, and to perform automated medical intervention. A textile-based fingertip oximeter is being created. There are wearable computer motherboards. A wearable camera with nano-optical fibers is being investigated.

Technology exists for a psoriasis treatment pajama. This could monitor plaque size and shape through a collated camera, simultaneously delivering laser light in the NBUVB range in a pattern which matches the patient's lesions.

Nanotechnology has created a host of novel technologies using nanomaterials. Nanoparticles can be extraordinarily and uniquely capable of drug delivery to the site of action. Novel pharmaceutical possibilities have been created. The FDA has already approved at least 12 products based on liposomes and/or other nanoparticles, including Onpattro, Doxil, Abraxane, Ontak, Toxol, and a home pregnancy test. Nanotechnology has the potential for a rapid test for bacterial resistance, as well as rapid diagnostic tests for dengue, syphilis, scabies, and leishmaniasis.

Cancer treatment can be done using siRNA, magnetic NP, nanobombs, plasmonic nanobubbles, and nanolasers. A single base pair anomaly can be detected. Nanosurgery can be done to delete SNPs. Surgery can be done on live cells with nanoneedles. Laser welding, tissue welding, and nanorobots are possible. Three-dimensional printing of live cells on a nanofiber matrix can be used for organ replacement. Artificial bone has already been created using this technology. Single molecules can be detected and surgically manipulated. Single bacterium can be detected. Clinical testing is cheaper, faster, and more accurate using nanomaterials.

Sentinal lymph node mapping can be done photo-acoustically using nanoparticles. Theranostics is a term for the use of multifunctional nanoparticles, which can perform several functions,

 

including locating tumors and diagnosing and treating them simultaneously.

Nanoparticles can be engineered to persist for weeks to months for long-term continuous monitoring of treatments and tumors.

We are familiar with nanosized zinc and titanium as they comprise sunscreens, which are translucent, because their nanosizes are below the wavelength of light. The U.S. government allows nanozinc and nanotitanium to pass as "generally accepted as safe," based on macro and micro zinc oxide and titanium dioxide. This is illogical, because they do not act the same in the nano form as in the micro and macro forms. A thousand tons of titanium dioxide and zinc oxide are produced annually. Seventy percent of titanium dioxide and 30% of zinc oxide sunscreens contain nanoparticles. Titanium dioxide nanoparticles can cause DNA damage[26] and adduct formation.[27] With exposure to UV light, free radicals are generated.[28] Titanium dioxide nanoparticle agglomerates have been observed in intracellular organelles.[29] Zinc oxide nanoparticles have the potential to damage DNA.[30] Sunscreen containing zinc nanoparticles applied to humans has been shown to increase serum levels of zinc.[31] UV exposure and extremity flexion increase the potential for zinc[31] and titanium nanoparticles to be absorbed through intact skin. There is a concern about inhaling nanoparticles in spray-on sunscreens.

Nanoparticles form protein complexes and evade immunologic defenses. Gadolinium is a nanoparticle, and it is responsible for nephrogenic systemic fibrosis.

Probably the first report of cancer likely due to nanoparticle environmental exposure was in 1775 by Percivall Pott. He reported scrotal cancer in chimney sweeps due to soot.

Exposure to indoor open cooking flames, which commonly occurs in developing countries, is harmful due to the inhalation of nanoparticles. Asbestos is a nanoparticle. Lung cancer can be caused by silica, coal dust, and smoke. Restaurant workers are at increased risk for cancer likely due to their exposure to cooking smoke. There was a report of several workers in a nanoparticle factory, working without adequate personal protective equipment, who rapidly became fatally ill from exposure to large amounts of the nanoparticles they were producing.

Adverse environmental effects due to nanoparticles have been documented. Flame retardant chemicals have been found in the arctic. Silver nanoparticles can penetrate the stratum corneum. They cause spinal abnormalities, cardiac arrhythmias, and respiratory distress in fish. Carbon nanoparticles cause platelet aggregation. This may explain how pollution causes cardiovascular disease. Nanoparticles can be taken up by sensory nerves and delivered to the CNS. Soil particles can be detected in the inguinal lymph nodes of individuals who routinely go barefoot (podoconiosis). Fullerenes are toxic to bacteria and fish.

Coated nanoparticles decrease toxicity. Biodegradable nanoparticles made of locally sourced non-toxic materials may be safer. Grapefruit, soybean oil, and chitin can be used. These are called "green nanoparticles." PLA and PLGA nanoparticles are biodegradable and FDA approved.

## COVID-19 Vaccine

Highly effective immunization can be achieved with nanoparticles.[32] The HPV vaccine is a nanoparticle vaccine which uses virus-like particles (VLP).[2] Inoculation directly into the lymph node with nanoparticles can produce a robust immune response.[33]

The prediction of a 50–80% effectiveness for a COVID vaccine was fortunately an underestimation. Biotechnology to develop COVID-19 vaccines is superior due to the rapidity of development, cost-effectiveness, increased safety, and ease of manufacturing.[34] Most importantly, there is a highly effective ability to tune the vaccines in response to immune-evading, rapidly mutating viral variants. Emerging technology is not always successful, however. Effective vaccines for HIV and RSV have not been created, even with huge NIH investments.

Herd immunity is predicted to develop when 70% of the world's population is immune (5.6 billion people). The Swedish experiment to allow natural infection to lead to herd immunity has proved disastrous.[8] This deadly pandemic be only be controlled and arrested with a safe and effective vaccine.[35,36] Bioinformatics, cheminformatics, AI, Big Data, the Internet of Things, and machine learning can be useful in vaccine development.

Traditional vaccine development takes 10–15 years[37,38] and is typically only 6% successful. The Ebola virus vaccine took 43 years to develop after the virus was identified. Novel uses of emerging technologies are currently available to create next-gen vaccines. Nanotechnology and intelligent rational vaccine design using reverse vaccinology have revolutionized vaccine development. Advanced immuno-informatic strategies[39] were implemented. In silico structural biology studies and molecular dynamics simulations were performed. Cooperation occurred among countries, governments, and NGOs, including U.S.'s WARP SPEED and NIH's accelerated COVID therapeutic interventions and vaccines (ACTIV),[40] WHO's COVAX[41] and COVID-19 tools accelerator (ACT), coalition for epidemic preparedness innovations (CEPI), and global alliance for vaccines and immunization (GAVI). They coordinated global vaccine development and promoted rapid evolutionary smart biomaterial vaccine development, encouraging umbrella trials.[42] This has not been done with the attempts to create Tb, malaria, or HIV vaccines. Novel technologies including a "molecular clamp" have been created.[2,8]

It is extraordinary that within 4 months of the viral genome being released, over 259 vaccines had been in development. At least four currently provisionally approved by governments were being given to millions of people within a year of the identification of the virus. MERS and SARS are also betacoronaviruses. The incomplete work done due to the essential resolution of

those epidemics was nonetheless crucial to the rapid development of COVID-19 vaccines.[45] One mRNA vaccine for COVID-19 was in clinical trials 66 days after the virus was sequenced. Adenovirus-based vaccines are being utilized.[46] Antibody treatment for COVID-19 is likewise currently available. Molecular farming uses genetically engineered plants to produce vaccines.[2,43,44] Other preventative and treatment modalities in development include nanobodies,[47] CRISPR-cas13a,[48] RNA, RNAi, miRNA,[50] DNA, subunit vaccines, mitochondrial targeted nanoparticles, therapeutic vaccines, nano-sponge,[2,49] virus-like particles, live attenuated virus, inactivated viruses, fusion-protein vaccines, recombinant protein vaccines, therapeutic antiviral peptides, ankara vector,[10] protein cages, ferritin nanoparticles, topical vaccines, and personalized cell therapy.[8]

Some of the "nano" lipoparticles in COVID-19 vaccines are reported to be larger than nanosized, up to 1000 nm.[51,52] Although the publications refer to them as nano, these are technically microparticles not nanoparticles. The larger size is likely to make them somewhat less effective than the true smaller-sized nanoparticles.

The diseasome and immunologic reactions to COVID-19 are complex. The main concerns for the ongoing monitoring of the current vaccines is the possibility of antibody-dependent enhancement (ADE)[41,53–60] or vaccine-dependent enhancement (VDE) (vaccine-induced immunopathology), phenomena whereupon protective antibodies cause worsening instead of prevention of disease. ADE is seen in dengue, when subsequent contagion with a different serotype can cause dengue hemorrhagic fever. A similar effect has been documented with Zika as well. The RSV vaccine was withdrawn in the 1960s due to ADE. ADE was also seen with the development of the SARS vaccine. ADE has already been described in COVID-19 infection per se.[18] ADE is the likely explanation for the unusual severity and variety of seemingly bizarre associated non-pulmonary findings and sequela of COVID-19, including autoimmunity. ADE is a purported cause of COVID vasculopathy, as antiphospholipid antibodies may be present. Antibodies can be protective or pathogenic. Partially effective or tapering levels of neutralizing antibodies can cause a deleterious increase in the attachment of the virus to the Fc receptor on the cell membrane creating a "Trojan horse effect," worsening viral disease by enhancing rather than preventing cell entry. The possibility of ADE or VDE exists as immunity fades or as the virus mutates. Immune complexes can occur with antibody targeted virus that is not completely cleared. Fortunately, so far, no ADE or VDE has been seen associated with the COVID vaccines. It is essential that there is monitoring for this potential phenomenon as the virus mutates and immunity tapers.

Antibody titers are easier to measure and have been touted in terms of neutralizing antibodies. However, it is the memory B and especially memory T cells which are more important in terms of a durable immunity to the virus.[8,41,61–63] Immunity induced by CD4 and CD8 T cells is less dominated by spike protein than in previous coronavirus infections. T cells can also amplify tissue damage due to an eosinophilic proinflammatory response. An immune reaction toward Th2,[59] as opposed to Th1,[2] leads to an increased risk for ADE.

The immensely successful development of COVID-19 vaccines has the potential to translate to a significant Moore's law like increase in the speed of development of further useful biotechnology resources for society. Rapid mutations occur in coronaviruses due to the low fidelity of the RNA polymerase.[17] Increasing immune pressure has already led to the SARS-CoV-2 virus evolving. COVID-19 is also predicted to become cyclically, seasonally endemic like yearly influenza.[2,64–66] New COVID-19 vaccines are already being developed to match novel variants exhibiting antigenic drift as is done with yearly flu vaccines. As opposed to inactivated or killed virus vaccines, mRNA and other bioengineered vaccines can rapidly and inexpensively be altered to match mutated antigenic epitopes. The technology will be effective if it is faster than the virus can mutate to evade it and a high enough percent of the population is immunized. The end of the current pandemic appears realistically within reach using a combination of nanomedicine technologies.

Bats, which are the most common reservoir for coronaviruses, harbor up to 5000 different viruses. SARS and SARS-CoV-2 are zoonotic bat viruses. It is inevitable that future zoonoses linked to bats will cause more coronavirus epidemics. As we have had SARS in 2002, MERS (from camels) in 2012, and now COVID-19, there is the inevitability of coronavirus and other emerging pathogen epidemics and pandemics, which will need to be dealt with in the future with further advancements in biotechnology and nanotechnology. Research is ongoing for a universal coronavirus vaccine.[67] Then we may finally be able to say we cured the common cold (those caused by coronavirus). Although it was certainly easier to go to the moon and Mars.

## References

1 Peiris M, Leung GM. What can we expect from first-generation COVID-19 vaccines? *Lancet* 2020; **396**: 1467–1469.
2 Machhi J, Shahjin F, Kevadiya BD. Nanocarrier vaccines for SARS-CoV-2. Adv Drug Deliv Rev 2021 https://doi.org/10.1016/j.addr.2021.01.002 [epub ahead of print].
3 Rinaldi A. RNA to the rescue: RNA is one of the most promising targets for drug development given its wide variety of uses. *EMBO Rep* 2020; **21**: e51013.
4 Mitchell MJ, Billingsley MM, Haley RM, *et al*. COVID-19 is an emerging, rapidly evolving situation. *Nat Rev Drug Discov* 2020; **4**: 1–24.
5 Nandedkar TD. Nanovaccines: recent developments in vaccination. *J Biosci* 2009; **34**: 995–1003.
6 Haynes BF. A new vaccine to battle Covid-19 [sp]. *N Engl J Med* 2020; NEJMe2035557.
7 Kaye AD, Cornell EM, Kevil CG, *et al*. Biology of COVID-19 and related viruses: epidemiology, signs, symptoms, diagnosis, and treatment. *Best Pract Res Anaesthesiol* 2020. PMC7723419 [ePub ahead of print].
8 Chung JY, Thone MN, Kwon YJ. COVID-19 vaccines: the status and perspectives in delivery points of view. *Adv Drug Deliv Rev* 2020. PMC7759095.

 

9 Balakrishnan VS. The arrival of Sputnik V. *Lancet Infect Dis* 2020; **20**: 1128.
10 Mishra SK, Tripathi T. One year update on the COVID-19 pandemic: where are we now? *Acta Trop* 2021; **214**: 105778.
11 Badgujar KC, Badgujar VC, Badgujar SB. Vaccine development against coronavirus (2003 to present): an overview, recent advances, current scenario, opportunities and challenges. *Diabetes Metabolic Syndrome: Clin Res Rev* 2020; **14**: 1361–1376.
12 Molaei S, Dadkhah M, Asghariazar V, *et al*. The immune response and immune evasion characteristics in SARS-CoV, MERS-CoV, and SARS-CoV-2: vaccine strategies. *Int Immunopharmacol* 2020. PMC7522676.
13 Zhou T, Su TT, Mundianto T. Immune asynchrony in COVID-19 pathogenesis and potential immunotherapies. *J Exp Med* 2020; **217**: e20200674.
14 Rendeiro AF, Casado J, Vorkas CK, *et al*. Profiling of immune dysfunction in COVID-19 patients allows early prediction of disease progression. *Life Sci Alliance* 2021; **4**: e202000955.
15 Usmani SS, Raghava GP. Potential challenges for coronavirus (SARS-CoV-2) vaccines under trial. *Front Immunol* 2020; **11**: 561851.
16 Wang X, Gui J. Cell-mediated immunity to SARS-CoV-2. *Pediatr Investig* 2020; **4**: 281–291.
17 Han HJ, Nwagwu C, Anyim O, *et al*. COVID-19 and cancer: from basic mechanisms to vaccine development using nanotechnology. *Int Immunopharmacol* 2021; **90**: 107247.
18 Haidere MF, Ratan ZA, Nowroz S, *et al*. COVID-19 vaccine: critical questions with complicated answers. *Biomol There (Seoul)* 2020; **29**: 1–10.
19 Koirala A, Joo YJ, Khatami A, *et al*. Vaccines for COVID-19: the current state of play. *Paediatr Respir Rev* 2020; **35**: 43–49.
20 Jain K. *The Handbook of Nano Medicine*. Totowa, NJ: Springer/Humana Press, 2012.
21 Prasad P. *Introduction to Nano Medicine and Nano Bioengineering*. Hoboken, NJ: Wiley, 2012.
22 Coetzee D, Venkataraman M, Militky J, *et al*. Influence of nanoparticles on thermal and electrical conductivity of composites. *Polymers (Basel)* 2020; **12**: 742.
23 Das A. Liquid drop model explaining melting point depression of nanoparticles. *Resonance* 2020; **25**: 215–231.
24 Rodriguez-Izquierdo I, Serramia MJ, Gomez R, *et al*. Gold nanoparticles crossing the blood-brain barrier prevent HSV-1 infection and reduce herpes associated amyloid-beta secretion. *J Clin Med* 2020; **9**: 155–156.
25 Wang L, Chen YP, Miller K. Functionalised [sp] nanoparticles complexed with antibiotic efficiently kill MRSA and other bacteria. *J Biomaterial Nanobiotechnol* 2014; **50**: 12030–12033.
26 Trouiller B, Reliene R, Westbrook A, *et al*. Titanium dioxide nanoparticles induce DNA damage and genetic instability in vivo in mice. *Cancer Res* 2009; **69**: 8784–8789.
27 Bhattacharya K, Davoren M, Boertz J, *et al*. Titanium dioxide nanoparticles induce oxidative stress and DNA-adduct formation but not DNA-breakage in human lung cells. *Particle and Fibre Toxicology* 2009; **6**: 17. https://doi.org/10.1186/1743-8977-6-17 [accessed on 25 January 2021].
28 Reeves JF, Davies SJ, Dodd NJF, *et al*. Hydroxyl radicals (OH) are associated with titanium dioxide ($TiO_2$) nanoparticle-induced cytotoxicity and oxidative DNA damage in fish cells. *Mutat Res* 2008; **640**: 113–122.
29 Lammel T, Mackevica A, Johansson BR. Endocytosis, intracellular fate, accumulation, and agglomeration of titanium dioxide ($TiO_2$) nanoparticles in the rainbow trout liver cell line RTL- W1. *Environ Sci Pollut Res* 2019; **26**: 15354–15372.
30 Attia H, Nounou H, Shalaby M. Zinc oxide nanoparticles induced oxidative DNA damage, inflammation, and apoptosis in rat's brain after oral exposure. *Toxics* 2018; **6**: 29.
31 Gulson B, McCall M, Korsch M, *et al*. Small amounts of zinc from zinc oxide particles in sunscreens applied outdoors are absorbed through human skin. *Toxicol Sci* 2010; **118**: 140–149.
32 Hanson MC, Crespo MP, Abraham W, *et al*. Nanoparticulate STING agonists are potent lymph node-targeted vaccine adjuvants. *J Clin Invest* 2015; **125**: 2532–2546.
33 Johansen P, Kundig TM. Intralymphatic immunotherapy and vaccination in mice. *J Vis Exp* 2014; **84**: e51031.
34 Wang F, Kream RM, Stefano GB. An evidence based perspective on mRNA-SARS-CoV-2 vaccine development. *Med Sci Monit.* 2020; **26**: e924700.
35 Lv H, Wu NC, Mok CK. COVID-19 vaccines: knowing the unknown. *Eur J Immunol* 2020; **50**: 939–943.
36 Wang J, Peng Y, Zu H, *et al*. The COVID-19 vaccine race: challenges and opportunities in vaccine formulation. *AAPS Pharm Sci Tech* 2020; **21**: 225.
37 Calina D, Docea AO, Petrakis D, *et al*. Towards effective COVID-19 vaccines: updates, perspectives and challenges (review). *Int J Mole Med* 2020; **46**: 3–16.
38 Jeyanathan M, Afkhami S, Xing Z. Immunological considerations for COVID-19 vaccine strategies. *Nat Rev Immunol* 2020; **4**: 1–18.
39 Joshi A, Joshi B, Amin-ul Mannan M, *et al*. Epitope based vaccine prediction for SARS- COV-2 by deploying immune-informatics approach. *Inform Med Unlocked* 2020; **19**: 100338.
40 Iserson KV. SARS-CoV-2 (COVID-19) vaccine development and production: an ethical way forward. *Camb Q Healthc Ethics* 2020; 1–10.
41 Alter G, Bingham K, Corey L., *et al*. interviewed by DeFrancesco L. Whither COVID-19 vaccines? Nature Biotechnology 2020; https://doi.org/10.1038/s41587-020-0697-7
42 Mullard A. COVID-19 vaccine development pipeline gears up. *Lancet* 2020; **395**: 1751–1752.
43 Rosales-Mendoza S, Marquez-Escobar VA, Gonzalez-Ortega O. Vaccines 2020; **8**: 183.
44 Rosales-Mendoza S. Will plant-made biopharmaceuticals play a role in the fight against COVID-19? Expert Opinion on Biological Therapy https://doi.org/10.1080/14712598.2020.1752177
45 Smith TR, Patel A, Ramos S, *et al*. Immunogenicity of a DNA vaccine candidate for COVID-19. *Nat Commun* 2020; **11**: 2601.
46 Kalita P, Padhi A, Zhang KY, Tripathi T. Design of a peptide-based subunit vaccine against novel coronavirus SARS-CoV-2. *Miicrob Pathog* 2020; **145**: 104236.
47 Ezzikouri S, Nourlil J, Tsukiyama-Kohara K, *et al*. Nanobodies: an unexplored opportunity to combat COVID-19. *J Biomol Struct Dyn* 2020.
48 Wang L, Zhou J, Kang C, *et al*. Rapid design and development of CRISPR-Cas13a targeting SARS-CoV-2 spike protein. *Theranostics* 2021; **11**: 649–664.
49 Witika BA, Makoni PA, Walker RB. Nano-biomimetic drug delivery vehicles: potential approaches for COVID-19 treatment. *Molecules* 2020; **25**: 5952.
50 El-Nabi SH, Elhiti M, El-Sheekh M. A new approach for COVID-19 treatment by micro-RNA. *Med Hypothesis* 2020; **143**: 110203.
51 Park KS, Bazzill JD, Son S, *et al*. Lipid-based vaccine nanoparticles for induction of humoral immune responses against

HIV-1 and SARS-CoV-2. *J Control Release* 2020. https://doi.org/10.1016/j.conrel.2020.12.031 [epub ahead of print].
52 Cavalvanti ID, de Britto C, Lira NM. Pharmaceutical nanotechnology: which products are [sp] been designed against COVID-19. *J Nanopart Res* 2020; **22**(9):276.
53 Ura T, Yamashita A, Mizuki N, *et al*. New vaccine production platforms used in developing SARS-CoV-2 vaccine candidates. *Vaccine* 2021; **39**: 197–201.
54 Karthik K, Maragatham T, Senthilkumar A, *et al*. Role of antibody-dependent enhancement (ADE) in the virulence of SARS-CoV-2 and its mitigation strategies for the development of vaccines and immunotherapies to counter COVID-19. Human Vaccines & Immunotherapeutics. https://doi.org/10.1080/21645515.2020.1796425
55 Peeples L. Avoiding pitfalls in the pursuit of a COVID-19 vaccine. *PNAS* 2020; **117**: 8218–8221.
56 Hotez PJ, Corry DB, Bottazzi ME. COVID-19 vaccine design: the janus face of immune enhancement. *Nature Rev Immunol* 2020; **20**: 347–348.
57 Wen J, Cheng Y, Ling R, *et al*. Antibody-dependent enhancement of coronavirus. *Int J Infect Dis* 2020; **100**: 483–489.
58 Morris K. The improbability of the rapid development of a vaccine for SARS-CoV-2. *Mol Ther* 2020; **28**: 1548e924700-e924700-8.
59 Bos R, Rutten L, Schuitemaker H. Ad26 vector-based COVID-19 vaccine encoding a prefusion-stabilized SARS-CoV-2 spike immunogen induces potent humoral and cellular immune responses. *NPJ Vaccines* 2020; **5**: 91.
60 Burton DR, Walker LM. Rational vaccine design in the time of COVID-19. *Cell Host Microbe* 2020; **27**: 695–698.
61 Haun BK, Lai C-Y, Williams CA. CoVaccine HT adjuvant potentiates robust immune responses to recombinant SARS-CoV-2 Spike S1 immunization. *Front Immunol* 2020; **11**: 599587.
62 Sauer K, Harris T. An effective COVID-19 vaccine needs to engage T cells. *Front Immunol* 2020; **11**: 581807.
63 Hellerstein M. What are the roles of antibodies versus a durable, high quality T-cell response in protective immunity against SARS-CoV-2? *Vaccine: X* 2020; **6**: 100076.
64 Perez de la Lastra JM, Baca-Golzalez V, Asensio-Calavia P, *et al*. Can immunization of hens provide oral-based therapeutics against COVID-19. *Vaccines* 2020; **8**: 486.
65 Kurup D, Wirblich C, Schnell MJ, *et al*. Rabies virus-based COVID-19 vaccine CORAVAX induces high levels of neutralizing antibodies against SARS-CoV-2. *NPJ Vaccines* 2020; **5**: 98.
66 Brown S, Brown T, Cederna PS, *et al*. The race for a COVID-19 vaccine: current trials, novel technologies, and future directions. *Plast Reconst Surg Glob Open* 2020; **8**: e3206.
67 Giurgea LT, Han A, Memoli MJ. Universal coronavirus vaccines: the time to start is now. *NPJ Vaccines* 2020; **5**: 43.

 