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# Carbohydrates-based diagnosis, prophylaxis and treatment of infectious diseases: Special emphasis on COVID-19

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## ABSTRACT

COVID-19 pandemic is taking a dangerous turn due to unavailability of approved and effective vaccines and therapy. Currently available diagnostic techniques are time-consuming, expensive, and maybe impacted by the mutations produced in the virus. Therefore, investigation of novel, rapid, and economic diagnosis techniques, prophylactic vaccines and targeted efficacious drug delivery systems as treatment strategy is imperative. Carbohydrates are essential biomolecules which also act as markers in the realization of immune systems. Moreover, they exhibit antiviral, antimicrobial, and antifungal properties. Carbohydrate-based vaccines and therapeutics including stimuli sensitive systems can be developed successfully and used effectively to fight COVID-19. Thus, carbohydrate-based diagnostic, prophylactic and therapeutic alternatives could be promising to defeat COVID-19 propitiously. Morphology of SARS-CoV-2 and its relevance in devising combat strategies has been discussed. Carbohydrate-based approaches for tackling infectious diseases and their importance in the design of various diagnostic and treatment modalities have been reviewed.

## 1. Introduction

Novel Coronavirus disease (COVID-19) is caused by severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) (WHO, 2020; Peng et al., 2020; Zhu et al., 2019). There are four main types of coronaviruses such as alpha ( $\alpha$ ), beta ( $\beta$ ), gamma ( $\gamma$ ) and delta ( $\delta$ ), among which only  $\alpha$ - and  $\beta$ -coronaviruses serve as pathogenic strains for humans (Cui et al., 2019). The primary COVID-19 symptoms include fever, severe respiratory problems, and pneumonia. The COVID-19 outbreak, across the whole world, has created many challenges in quick diagnosis, prophylaxis and cure of this dangerous infection. The widely accepted and commonly used diagnostic test i.e. real-time polymerase chain reaction (RT-PCR) is tedious and expensive. Furthermore, no amalgamated standard treatment strategy including vaccines, antibodies (Abs), and drugs against SARS-CoV-2 has been discovered yet. Presently, scientists are trying to develop multipurpose vaccines which stimulate the immune system to start producing Abs capable of neutralizing the virus. However, the genomic and proteomic diversities of SARS-CoV-2 are becoming a challenge for vaccine development (Liu et al., 2020).

Coronaviruses are enveloped single-stranded positive-sense RNA viruses containing spike glycoprotein (S-protein) that assumes crucial

role in the pathogenesis of coronavirus, and inducing host immune responses (Anthony et al., 2017; Liu et al., 2020). The S-protein of SARS-CoV-2 is occupied by 66 glycosylation sites, each of which can be occupied by up to 10 different glycans (Freeman et al., 2020). These S-protein and glycans (carbohydrates) are assumed to be chief targets in the development of new diagnostic techniques, as well as primary (vaccines) and secondary (small molecule drugs or biologics) treatment strategies (Freeman et al., 2020; Liu et al., 2020).

The carbohydrate based biomolecules are reported to be less toxic, biocompatible and immunogenic in traditional Chinese herbal medicine. The synthetic glycan molecules mimicking receptor of the viruses are found promising in the diagnosis of viruses (Hieu et al., 2014). Both plant and micro-organisms derived polysaccharides such as chitosan (CS), glucans, dextran, inulin, etc. have shown immunogenic behavior by stimulation of both antibody-based and cellular immunity therefore these are used as adjuvants in the development of vaccines (Nikolai & Peter, 2011). Large number of carbohydrates such CS, alginate and carrageenan (CRG) are found to be effective against diverse viruses and used as antiviral agents. Besides, several glycan-based molecules as antiviral drugs are in the clinical trial stages (Morokutti-Kurz, Graf & Prieschl-Grassauer, 2017). Thus, present review is focused to summa-

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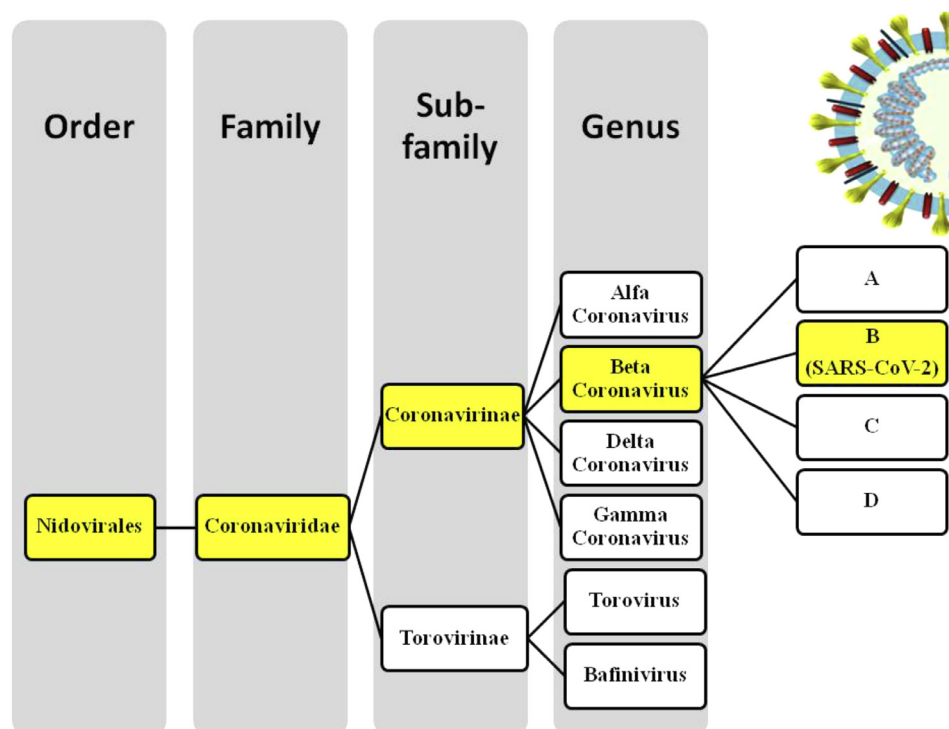


Fig. 1. Classification of coronaviruses (Thanigaimalai et al., 2020).

size the existing knowledge (available in the literature database) pertaining to the carbohydrate-based diagnostic strategies, antiviral properties of carbohydrates and carbohydrate-based drug molecules, applications of carbohydrates as adjuvants in vaccines, and carbohydrate-based nanoparticles as vaccine and drug carriers for more infectious diseases caused by other viruses. In the end, these potential strategies are correlated to SARS-CoV-2.

## 2. Structure of SARS-CoV-2

Coronaviruses are mainly categorized in order *Nidovirales* under the family *Coronaviridae* (CRVD). The two sub-families of CRVD include *Toroviruses* and *Coronaviruses*. The further classification, on the basis of rooted and unrooted genetic trees and partial nucleotide sequence of RNA-dependent RNA polymerase, into four genera include alpha ( $\alpha$ ) coronavirus, beta ( $\beta$ ) coronavirus, gamma ( $\gamma$ ) coronavirus, and delta ( $\delta$ ) coronavirus (Fig. 1). The first two ( $\alpha$ - and  $\beta$ -coronaviruses) mainly infect warm-blooded animals, while the later two ( $\gamma$  and  $\delta$ -coronaviruses) infect birds, and few of them can also infect mammals (Thanigaimalai, Sangeetha & Manoj, 2020).

SAR-CoV-2 is a single-stranded, enveloped, non-segmented virus having a diameter ranging from 65 to 125 nm, with the outer surface showcasing crown-like spikes (Huang et al., 2020; ul Qamar, Alqahtani, Alamri & Chen, 2020). Amongst a total of 380 amino acid substitutions, 27 substitutions present in the spike glycoprotein (S-protein) of SARS-CoV-2 play an imperative role in viral entry into the host cell and can be responsible for antibody-mediated virus-neutralization (Kumar, Maurya, Prasad, Bhatt & Saxena, 2020; Wu et al., 2020). The different structural proteins of betacoronaviruses (Fig. 2) are spike protein (S), an envelope protein (E), membrane protein (M), and nucleocapsid protein (N) (Jiang, Hillyer & Du, 2020). The spike protein contains 22 probable N-linked glycosylation and 4 probable O-linked glycosylation sites. Besides, the protein has two useful subunits specifically S1 and S2. The subunit S1 is associated with receptor binding and subunit S2 is responsible for membrane fusion (Guo et al., 2020). In SARS-CoV-2, though there is a structural resemblance with SARS-CoV, the receptor-binding domain for binding to angiotensin-converting enzyme (ACE-2) differs

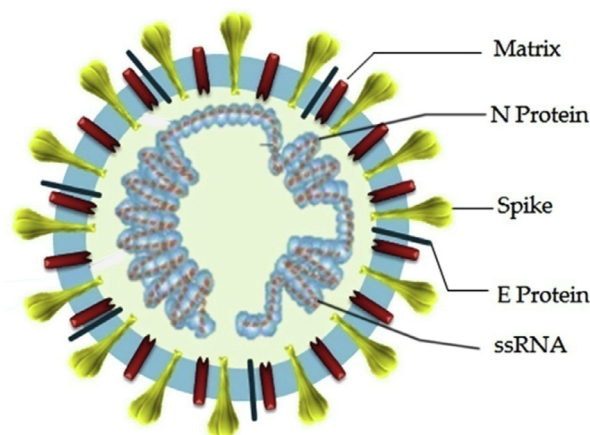


Fig. 2. Structure of SARS-CoV-2.

in five of the existing six amino acid residues (Andersen, Rambaut, Lipkin, Holmes & Garry, 2020). There is an absence of cross-reactivity in monoclonal antibodies (mAbs) meant for SARS-CoV-S receptor binding sites and SARS-CoV-2-spike glycoprotein (SARS-CoV-2-S) receptor binding sites (Wrapp et al., 2020). The surface of the envelope spike is mainly dominated by the glycans derived from the host. The viral pathology such as mediating protein folding and stability is basically affected by the viral glycosylation (Watanabe, Bowden, Wilson & Crispin, 2019). The spike protein is of chief consideration in designing the vaccine. Moreover, recombinant viral spike glycosylation also provides valuable information in vaccine design (Amanat & Krammer, 2020; Behrens et al., 2017; Cao et al., 2017).

## 3. Carbohydrates-based diagnosis of COVID-19

The unexpected flare-up of COVID-19, globally, has created a massive burden and challenges in the rapid diagnosis of SARS-CoV-2. Currently, the polymerase chain reaction (PCR) is the most widely used

technique for COVID-19 diagnosis; however, a laboratory setting is necessary for it. Besides, it is tedious and expensive. Furthermore, the genetic code of the virus is changing (mutating) during its development thereby limiting the effectiveness of the PCR test (Forouzes, Rahimi, Valizadeh, Dadashzadeh & Mirzazadeh, 2020). The various types of mutations in SARS-CoV-2 reported by different studies include missense, synonymous, insertion, deletion, and non-coding mutations (Chan et al., 2020; Chen, Li, Li, Ren & Hu, 2020; Koyama, 2020; Mercatelli & Giorgi, 2020; Wang et al., 2020; Yin, 2020). Of these, the missense and synonymous mutations were the most common forms of mutations along the SARS-CoV-2 genome (Chan et al., 2020; Laamarti et al., 2020). The kinds of mutations observed in different genes of SARS-CoV-2 involved accessory proteins (ORF1ab, ORF3a, ORF6, ORF7, ORF8, ORF10) S, M, E, and N (Laamarti et al., 2020; Mercatelli, Giorgi, 2020; Coronavirus disease (COVID-19): Virus evolution 2020). Besides this, large numbers of mutations are reported in the genes such as non-structural proteins (nsp1, nsp2 nsp3, nsp12, and nsp15) of ORF1ab, S, and ORF8 compared to other genes (Chan et al., 2020; Koyama, 2020; Laamarti et al., 2020). In a current study, ten hotspot mutations including D614G (23403A>G) on S, L84S (28144T>C) on ORF8, S5932F on nsp14, M5865V on nsp13, L37F (10818G>C) on nsp6, T85I (1059C>T) of nsp2, Q57H (25563G>T) on ORF3a, G251V (26144G>T) on ORF3a, R203K (28881G>A) on N, as well as G204R (28883G>C) on N with a frequency of over 0.10 were reported in SARS-CoV-2 genomes (Laamarti et al., 2020). Furthermore, recently, many countries have reported different variants of SARS-CoV-2 to World Health Organization (WHO) (SARS-CoV-2 variants 2020). The SARS-CoV-2 VOC 202012/01 variant observed in United Kingdom (UK) has shown 23 nucleotide substitutions. SARS-CoV-2 VOC 202012/01 from the UK has also displayed N501Y mutation. In addition, the deletion at the position of 69/70del is another mutation found in the VOC 202012/01 variant. In another case, the variant undergoing N501Y mutation was observed in South Africa and is named as variant 501Y.V2. Similarly, the variant 501Y.V2 also carries other mutations named as chE484K and K417N. To date, the above variants have been also reported in many other countries/territories/areas in five of the six WHO regions (WHO, 2020). Studies conducted by numerous scientists suggested that these variants are more transmissible (spread easily and quickly) than other variants, which may lead to more cases of COVID-19. However, they would not cause more severe illness or higher mortality or any kind of distinct clinical manifestations (WHO, 2020).

These mutations can influence the sensitivity of the available diagnostic techniques (RT-PCR: non-carbohydrate based technique). Therefore, there is a dire need to investigate novel and rapid COVID-19 diagnostic techniques wherein the genetic mutations do not influence the sensitivity and rapidity of the diagnosis test.

It is observed that the viral S-protein and the host ACE-2 receptor are widely glycosylated upon infection. After the attack of viruses in the human body through the respiratory tract, they usually utilize sugar chains (glycans) present on the surface of host cells. It is found that viral S-protein contains 66 glycosylation sites, each of which can be occupied by up to 10 different glycans when tested (A representative illustration presented in Fig. 3). Thus, the virus is covered by glycans that are resistant to mutation through its development process. Therefore, the diagnosis will stay convincing even if there is any mutation in the genetic code (Freeman et al., 2020; Watanabe, Allen, Wrapp, McLellan & Crispin, 2020).

María and co-workers have developed a glycan-based technique for the detection of human influenza virus X31 (H3N2). They designed glycananoparticles (AuNPs functionalized with thiolated trivalent  $\alpha$ -2,6-thio-linked SA derivative) and successfully detected the H3N2 virus (María et al., 2013). Recently, Icen Diagnostics has come up with COVID-19 diagnostic approach by targeting sugar chains (glycans) present over the surface of SARS-CoV-2 and these glycans remains unchanged during mutation (Freeman et al., 2020; Watanabe et al., 2020). Icen Diagnostics have explained that the carbohydrate molecules cho-

sen in their diagnostic kit are specifically recognized by the target pathogens (influenza, COVID-19, etc.) and help to spot the virus. This diagnostic technique is under clinical trials currently. The Icen Diagnostics kit is very simple like a self test kit to detect pregnancy and indicates the presence of the viral infection in a sample obtained by taking a nasopharyngeal swab in around 15 min (Sophia, 2020). Thus, the Icen technology provides an accurate and rapid lateral flow test that detects the interaction of the virus with sugars across human cells. Besides, the kit will be very efficient due to the robustness of the results (Richardson, 2020). Moreover, Professor Matt Gibson and group (University of Warwick) are also developing a novel diagnostic kit in collaboration with Icen Diagnostics. The main components of their test kit include sugar (glycan) placed on the plane of paper or on gold nanoparticles. The main working principle of this test is that it will bind the sample with sugars present on the paper surface encasing gold NPs to produce a red line, demonstrating positive results, in the presence of the virus (Gibson, 2020).

Several previous studies have also utilized carbohydrates (glycan) for the detection of viruses. Hieu and co-workers have developed a range of synthetic glycans as receptor mimics of the virus and then printed these glycan channels onto a commercial glass slide via a free amine at the end of a spacer to produce a small focused microarray, and utilized it for the detection of viruses. This microarray has detected different strains of the influenza A virus (Hieu et al., 2014).

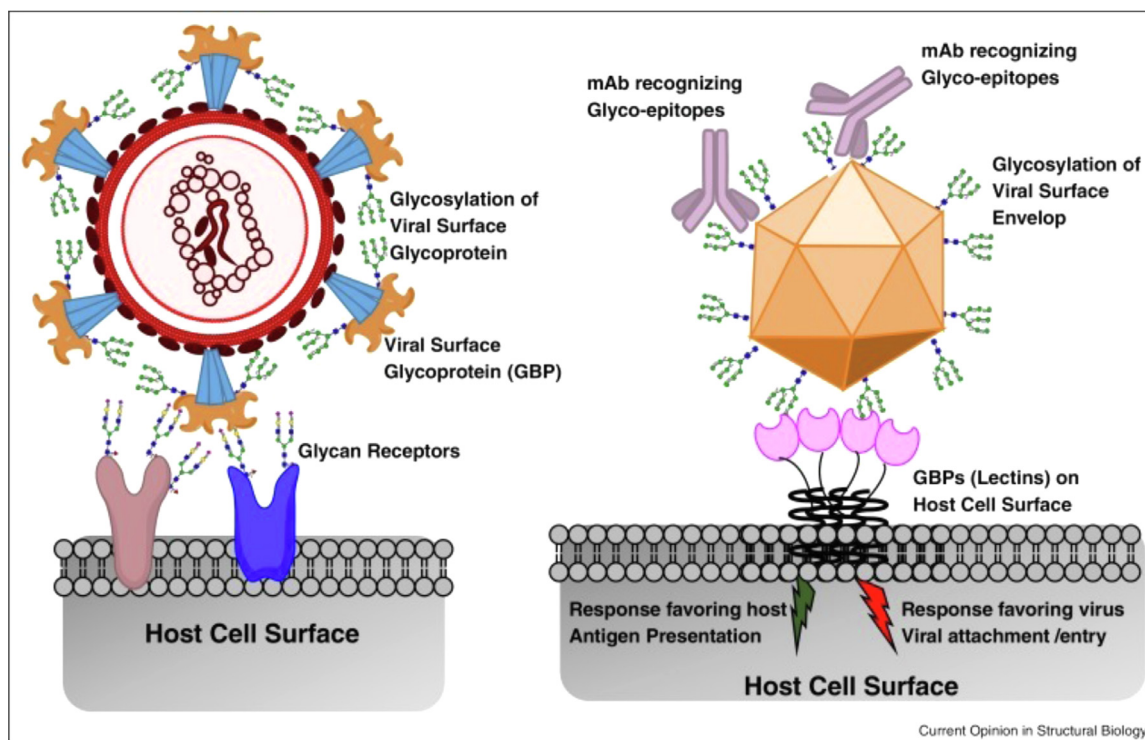
#### 4. Carbohydrates-based vaccine strategies

##### 4.1. Carbohydrates and their immunogenicity

Carbohydrates are generally found on the surface of bacteria, fungi, and viruses. Commonly, the completely structured and matured virus (virion) is decorated by the glycans (sugar coat) of the corresponding host and which is responsible for the escape of the virus from host immune recognition. In the case of the SARS-CoV-2, S-protein is heavily decorated by N-linked glycans projected from the surfaces of the virions (Yan et al., 2020). The coronavirus (CoV-S) glycans veil the protein surface and restrict antibody access to protein-neutralizing epitopes (Walls et al., 2016). Similarly, several other viruses such as human immunodeficiency virus (HIV-1) (Stewart-Jones et al., 2016), Lassa virus (LV) (Sommerstein et al., 2015), and hepatitis C virus (HCV) (Falkowska, Kajumo, Garcia, Reinus & Dragic, 2007) have shown N-linked glycans casing the uncovered protein surfaces like virus-neutralizing protein epitopes.

The carbohydrates serve as a chief category of viral antigens which are easily identified by the host immune system that stimulates the production of Abs (Katharina & Thierry, 2020). Thus, carbohydrates are called immunogen or antigen because they are able to induce as well to increase the immune response in the host. The immunogenic carbohydrate biomolecule is found to be a promising target in the development of vaccines for different types of infectious diseases (Ezzel, Abshire, Little, Lidgerding & Brown, 1990; Schneerson et al., 1980; Wang et al., 2007). Several polysaccharides such as glucans, dextran, inulin (fructans), etc. which are obtained from plants or micro-organisms demonstrated stimulation of both antibody-based and cellular immunity (Nikolai & Peter, 2011). Furthermore, the red algae-derived sulfated-carrageenan displayed a potent effect on the tobacco mosaic virus (TMV) infection by enhancing plant immunity (Dang et al., 2009). Another study, in animals and humans, indicated that CS stimulates both humoral and cell-mediated immune responses and also demonstrates a safety record. Therefore, CS has been used as an adjuvant for improving vaccine efficacy by enhancing immune response, particularly in RNA virus vaccines (Zaharoff, Rogers, Hance, Schlom & Greiner, 2007). Moreover, carbohydrates conjugated with carrier proteins and utilized for the vaccine productions resulted in improved immunogenicity (Shuyao & Xuefei, 2020). Based on the above observations, the carbohydrate moieties with immunogenic properties can be used as an ad-





**Fig. 3.** Schematic representation of complex glycans in the interplay between virus and host. Shown on the left is a schematic of a virus surface glycoprotein that recognized glycans on the host cell surface as their primary receptors for viral attachment and entry. The viral surface protein is in itself glycosylated and depending on the site of glycan occupancy, the glycosylation would impact the binding of this protein to the host-cell glycan receptor. Shown on the right is a graphic of glycan on the surface of virus envelop proteins (such as dengue) interacting with GBP anchored on the host cell. This interaction could either be beneficial for the virus wherein it plays a role in viral attachment and entry into a cell capable of promoting the productive infection or it could be beneficial to the host wherein antigen presenting cells could uptake the virus and prime the host immune response.

(Figure reprinted from Rahul et al., Glycan-protein interactions in viral pathogenesis, *Current Opinion in Structural Biology*, 40, 153–162, Copyright (2016) with permission from Elsevier). (Rahul et al., 2016)

juvant in the development of primary prophylactic strategies (vaccines) against numerous viral infections including COVID-19.

#### 4.2. Viral glycan shields as vaccine targets

Innovative strategies are critically expected to drive development of vaccines against novel viral infections. Glycosylation of viral glycoproteins has been depicted in detail in Fig. 3. Viruses lack glycosylation machinery required for glycan synthesis, and depend on host glycosylation machinery for the same, thereby expressing conserved carbohydrates of the virus. Furthermore, several glyco-antigens co-expressed by these viruses have also been recognized. These expressed conserved glyco-determinants may serve as appropriate broad-spectrum virus neutralization epitopes (Roberto & Sandro, 2020).

The S-protein of CoVs is mainly furnished by N-linked glycans jutting from the virion surfaces. The S-protein of SARS-CoV-2 (SARS-CoV-2-S) is composed of 22 N-linked glycosylation sites out of which 16 sites are observed to be glycosylated. Of the 22 sites, 20 N-linked glycosylation sites are conserved in SARS-CoV-2-S. Moreover, out of 13 sites, 9 sites in the S1 subunit, and all 9 sites in the S2 subunit are conserved among SARS-CoV-2-S and SARS-CoV-S (Walls et al., 2019). Thus, S-protein of SARS-CoV-2 would be a key target in the development of vaccine.

#### 4.3. Designing carbohydrate-based vaccines

There are currently more than 50 COVID-19 vaccine candidates in trials. Recent vaccines approved by India for emergency use include Covaxin (BBV152) and Covishield. The ChAdOx1 nCoV-19 vaccine (AZD1222) of Oxford/AstraZeneca, UK showed an efficacy of 67%

with no hospitalizations in the vaccine group 22 days after the first dose and 82% at the second dose given on 12 weeks or more after the first dose in phase III trials. COVID-19 Vaccine Moderna (mRNA-1273) of the USA and US government displayed 94% efficacy in phase III trial. Moreover, Pfizer-BioNTech COVID-19 Vaccine (BNT162b2) indigenous to the USA and Germany have demonstrated 95% efficacy in phase III trial. Furthermore, Russia has approved Sputnik V or Gam-Covid-Vac and interim analysis of phase III trial of this vaccine displayed 92% efficacy.

Covaxin is a whole-virion inactivated SARS-CoV-2 vaccine formulated with a Toll-like receptor (TLR) 7/8 agonist molecule adsorbed to alum (Algel-IMDG). Algel-IMDG is an imidazoquinoline molecule chemisorbed on alum (Algel) has been designed to transfer vaccine antigen directly to draining lymph nodes without diffusing into the systemic circulation (Raches et al., 2021). Covishield vaccine is prepared from a weakened version of adenovirus (common cold virus) from chimpanzees (WHO). The ChAdOx1 nCoV-19 vaccine (AZD1222) is composed of a replication-deficient chimpanzee adenoviral vector ChAdOx1, containing the SARS-CoV-2 structural surface glycoprotein antigen (S-protein; nCoV-19) gene (Merryn et al., 2021). COVID-19 Vaccine Moderna (virus-free vaccine) contains a molecule called messenger RNA (mRNA) with instructions to generate a protein from SARS-CoV-2 (European Medical Agency). BNT162b2 is a nucleoside-modified RNA lipid nanoparticle-formulated vaccine that encodes a prefusion stabilized, membrane-anchored full-length SARS-CoV-2-S (Fernando et al., 2020). Sputnik V or Gam-Covid-Vac is a human adenoviral vector-based vaccine composed of a recombinant adenovirus type 26 (rAd26) vector and type 5 (rAd5) vector that carries the gene of SARS-CoV-2-S (Denis et al., 2020).

**Table 1**  
Carbohydrate-based vaccines developed against different viruses.

| Indication  | Vaccine  | Significance   |
|---|--|--|
| Avian infectious bronchitis virus (AIBV)                            | Astragalus polysaccharides adjuvant-based AIBV vaccine                       | Improved both cellular and humoral immune response (Pengju et al., 2017).  |
| H3N2 influenza virus  | Polysaccharide (Advax™)-based trivalent inactivated influenza vaccine        | Increased immunogenicity with no safety issues at reduced dose of vaccine in men (David et al., 2016).   |
| Severe acute respiratory syndrome-associated coronavirus (SARS-CoV) | Delta inulin (polysaccharide adjuvant)-based inactivated whole-virus vaccine | Significantly enhanced serum neutralizing-antibody (nAb) titers and decreased lung virus titers in the presence of inulin. Enhanced and long-lived T-cell immunity was achieved in presence of delta inulin as adjuvant, thereby overcoming the natural tendency for rapidly waning coronavirus immunity (Yoshikazu et al., 2015). |
| SARS-CoV  | Inulin and CpG oligonucleotide-based inactivated SARS-CoV vaccine            | Significantly improved serums nAb titers lacking of lung immunopathology (Honda-Okubo et al., 2015).   |
| Hepatitis B virus   | Polysaccharide (Advax™)-based hepatitis B vaccine                            | Significantly improved cellular and humoral immune response in mice and guinea pigs (Fadi, Yoshikazu, Samay & Nikolai, 2013).  |
| Japanese encephalitis virus   | Japanese encephalitis vaccine (JEV) with inulin (adjuvanted vaccine)         | The inulin-based JEV vaccine displayed superior immunity in mice compared to non-adjuvanted vaccine (Mario et al., 2010).  |

WHO chief scientist Soumya Swaminathan recently mentioned that the knowledge of SARS-CoV-2 is still evolving (Soumya, 2021). Most scientists believe that the protection against different variants can be offered by the vaccines currently in development and a few of which have been approved since these vaccines evoke a fairly broad immune response (host of antibodies and cell-mediated immune responses). At present, large numbers of studies are going on in laboratories around the world to assure the effectiveness of these vaccines against new variants of the SARS-CoV-2. And there is a chance that these vaccines may be less effective against currently existed variants (SARS-CoV-2 VOC 202012/01 and 501Y.V2) (WHO, 2021). Besides, the rationale combination of these approved vaccines could be effective against these variants. However, further investigations are needed in this direction.

Advances in the glycosciences have created a new platform in the design of carbohydrate-based vaccines as potential alternatives to vaccines based on above approaches. The glycoproteins expressed on the virus body surface are mainly associated with immune evasion. Besides, the pathogenic viruses contain polysaccharides or lipopolysaccharides on their cell-surface that protect the virus from immune system of the host and prevent antibody-triggered complement activation and phagocytosis. Thus, the Abs that exclusively target viral surface polysaccharides enhances pathogen neutralization and elimination. The glycan structures present on the surface of pathogens including viruses and aberrant glycosylation are the chief targets to devise carbohydrate-based vaccines (Liangwei et al., 2020; Alexander et al., 2005). The carbohydrate-based vaccines developed against different types of viruses are depicted in Table 1.

Yoshikazu and group have developed a safe and effective new delta inulin (polysaccharide adjuvant)-based inactivated whole-virus vaccine against severe acute respiratory syndrome-associated coronavirus (SARS-CoV). This vaccine has displayed enhanced T-cell gamma interferon (IFN- $\gamma$ ) recall responses that result in the production of high neutralizing-antibody (nAbs) titers. In their study, they concluded the importance of the use of carbohydrates as an adjuvant in the development of effective vaccines against different viruses (Yoshikazu et al., 2015). In another study, David et al. have explored carbohydrate-based subcutaneous vaccination using CS adjuvant. They observed improved antigen-specific antibody titers over 5-folds and antigen-specific splenic CD4<sup>+</sup> proliferation over 6-folds in the presence of CS (David, Connie, Kenneth, Jeffrey & John, 2007). Similarly, Auwalu and co-workers have studied vaccination with Astragalus (containing mannose, xylose, D-glucose, and galactose) and Ginseng polysaccharides in chickens against H5N1 Avian Influenza Virus (H5N1 AIV). The obtained results demonstrated a remarkable immune response against H5N1 AIV revealing the potency of Astragalus and Ginseng polysaccharides as adjuvants in the development of the H5N1 AIV vaccine (Auwalu, Sanpha, Xingang, Yongliang & Guoqing, 2016). A CS-based Norovirus antigen vaccine designed by Zhao et al. has showed significantly enhanced potency of virus

nAbs due to the presence of CS polysaccharide (Zhao et al., 2020). However, the rapid gene mutations in the virus can cause alteration in glycosylation sites and also boost the structural diversity of viruses. Despite these challenges, the development of carbohydrate-based antiviral vaccines, for HIV and influenza viruses is much progressed which affirms a positive outlook for development of vaccines against numerous other viral infections.

Both polysaccharide vaccines (PSV) and glycoconjugate vaccines (GCV) can be the potential candidates to cater against several types of viral infections. The large number of similar antigenic epitopes present in the PSV causes activation of B-lymphocytes (B-cells) by interaction with the antigenic receptors (Anna-Karin & Carole, 2019; Vos, Lees, Wu, Snapper & Mond, 2010). These B cells can produce huge quantities of IgM antibodies (immunoglobulin) against polysaccharide antigens (Anna-Karin & Carole, 2019). In any case, the fundamental disadvantage of PSV is the production of peculiar immunity. The GCV prepared by conjugation of polysaccharides with immunogenic carrier proteins are able to overcome this drawback. Bernardes et al. have investigated the GCV vaccine against HITB (Adamo et al., 2013; Francesca et al., 2019). The vital findings from the previous studies described herein might be directly applied for the development of carbohydrate-based vaccines against SARS-CoV-2.

In the case of the SARS-CoV-2, the S-protein is known to be masked by glycans that promote the virus entry into the host cell. These S-proteins serve as the main immunogen and a key target of nAbs, forming an area of focus for vaccine development (Denong, 2020). The use of polysaccharides that imitate true COVID-19 polysaccharides on the S-protein would be of vital importance in the development of the effective carbohydrate-based vaccines. However, a diversified sequence of S-protein variants reported with mutations at more sites is a big challenge in the development of efficacious vaccines. In addition, the vaccines produced might not be efficient for future strains and we must be ready for the next outbreak. A versatile and robust strategy would be a mandate in order to deal with a pandemic of this stature in coming years.

#### 4.4. Carbohydrate-based nanocarriers (NCs) for vaccine development

Nanocarriers have recently gained attention as a promising tool in the development of new generation of vaccines because different NCs can accommodate the antigens and also act as an adjuvant in numerous cases. The different carbohydrate-based polymeric NCs and inorganic NCs that are surface coated with polysaccharides have been developed for antigen or vaccine delivery against diverse viruses (Pei, Felicity, Lisbeth, Zhi & Li, 2020; Shin et al., 2020). These NCs offer various benefits such as maximum payloads, improved protection against premature degradation and stability of antigen, increased immunogenicity, targeting ability, sustained release, adjustable sizes, as well as versatile

**Table 2**  
Carbohydrate-based NC vaccine (NCV) against different viral infections.

| Nanoparticles (NPs) used as vaccine carriers         | Indication   | Vaccines/Antigens                          | Significance  |
|--|--|--|---|
| Chitosan (CS)/alginate NPs                           | Porcine reproductive and respiratory syndrome (PRRS) | Bee venom                                  | Intranasal administration of bee venom CS/alginate NPs displayed significantly increased Th1-related responses including a high population of CD4 <sup>+</sup> T lymphocyte and improved virus clearance activity in the pig (Lee et al., 2018).                                    |
| Poly-ε-caprolactone (PCL)/ CS NPs                    | Hepatitis B  | HBsAg                                      | Increased immune response on intranasal administration of NPs in mice due to immunostimulatory and mucoadhesive effect of CS, and enhanced antigen stability due to the resistance of PCL against degradation in biological fluids (Jesus, Soares, Costa, Borchard & Borges, 2016). |
| N-trimethylaminoethylmethacrylate chitosan (TMC) NPs | Swine flu  | Influenza A (H1N1) antigen                 | The TMC NPs produced considerably higher levels of serum IgG and mucosal sIgA than free antigen (Liu et al., 2015).   |
| Chitosan NPs   | Hepatitis B  | Hepatitis B surface antigen (HBsAg)        | Significantly improved immunity in mice was noticed (Amit et al., 2013).  |
| Chitosan (CS) NPs                                    | Newcastle disease                                    | Live Newcastle disease virus (NDV) vaccine | NDV-CS-NPs vaccine demonstrated superior immune responses and protection against NDV compared to the live and inactivated NDV vaccines (Zhao et al., 2012).   |
| Chitosan NPs   | Swine flu  | DNA vaccine                                | Improved immune response and prolonged release of plasmid DNA from DNA vaccine encapsulated CS NPs than plain DNA vaccine (Kai et al., 2011).   |

surface characteristics (Vijayan, Mohapatra, Uthaman & Park, 2019). Another imperative attribute of NPs is that it can imitate parts of the original protein and activate the immune response through interactions with receptors of host cells (Gaurav et al., 2020; Poon et al., 2017). In addition, various polymeric NPs have showed a significantly increased immunogenic response of negatively charged DNA vaccines delivered using positively charged carbohydrate polymers such as CS (Shin et al., 2020). For instance, Kai and co-workers have developed a DNA vaccine encapsulated in CS NPs against swine influenza caused by the influenza virus. These CS NPs containing DNA vaccine exhibited notable immune response in mice due to the presence of CS (Kai et al., 2011). Protein antigen-loaded polysaccharide-coated calcium phosphate nanoparticles vaccine with improved oral stability has been fabricated by Pei and group. These NPs protected the antigens from acidic degradation in the gastrointestinal tract and exhibited improved immune response in the small intestine due to coating with CS and alginate polysaccharides (Pei et al., 2020). Nanocarrier vaccine (NCV) with targeting ability towards antigen-presenting cells is a modern approach investigated with the objective of enhancing host immunity (Aikins, Bazzill & Moon, 2017; Kim, Kye & Yun, 2019).

A new strategy of decorating nanovaccines with immune cell-targeting molecules such as carbohydrates can be devised to target antigen-presenting cells (dendritic cells). Rezaei and group have developed hepatitis B antigen (HBsAg)-loaded iron oxide nanoparticles (IONP); surface decorated with mannose (ligand) to target mannose receptor on dendritic cells. A remarkable immune response by the production of Abs against antigen was observed from targeted nanovaccine containing mannose compared to non-targeted nanovaccine (Rezaei, Hosseini, Khavari-Nejad, Najafi & Mahdavi, 2019). Some carbohydrate-based NCVs developed with improved performance are summarized in Table 2.

## 5. Antiviral property of carbohydrates

Carbohydrates exhibit promising biological activities including antiviral, antimicrobial (AMB), antifungal, and anti-inflammatory (Bezerra et al., 2018; Gao et al., 2019; Guiping et al., 2019; He, Zhang, Zhang, Linhardt & Sun, 2016). Amongst the carbohydrates, various marine polysaccharides such as CS, carrageenan (CRG), and their derivatives show good inhibitory activity against diverse viruses. Lozenges prepared using Iota-carrageenan have displayed antiviral activity against different viruses including human rhinovirus (HRV), influenza virus A H1N1 and H7N9, and human coronaviruses (HCoV), and are found to be promising therapy alternatives against viral infections of the throat (Mei et al., 2016; Morokutti-Kurz et al., 2017). Moreover, the

N-(2-hydroxypropyl)-3-trimethylammonium chitosan chloride (HTCC), cationically modified CS derivative (CM-HTCC) has showed momentous inhibition against the human coronaviruses such as HCoV-229E, HCoV-OC43, HCoV-NL63, and HCoV-HKU1. Similarly, hydrophobically modified HTCC (HM-HTCC) derivative is a powerful inhibitor of the coronavirus HCoV-NL63, signifying that modified CS derivatives are efficient inhibitors of all low-pathogenic human coronaviruses (Milewska et al., 2016). Furthermore, CRG nasal spray showed noteworthy antiviral efficacy against HRV, HCoV, and influenza A virus (IAV). Compared to other viruses, the utmost effectiveness was observed in human corona virus-infected patients (Koenighofer et al., 2014).

Modified polysaccharides such as sulfated polysaccharides possess strong polyanionic properties and can block the positive charge on the virus-cell surface to prevent virus adsorption or invasion (Chen, Huang, 2018). Furthermore, these polysaccharides interact with the surface of the virus by a negative charge that results in the inhibition of infection inducing ability of the virus. It has been proven that high molecular weight sulfated polysaccharide (Iota-carrageenan) has antiviral property and interacts directly with the viral surface (Morokutti-Kurz et al., 2017). In another study, Xiaofei et al. have synthesized the amino (6-amino-6-deoxy) derivative of CS with improved solubility. This modified CS showed significantly improved antiviral activity against bird Newcastle disease virus (causative agent of Newcastle disease) (Xiaofei et al., 2019).

The α-glucan-based standardized mushroom extract obtained from cultured *Lentinula edodes* mycelia showed immunostimulant activity in HIV, AIV, human papillomavirus (HPV), and hepatitis B virus (HBV) infected humans and animals by promoting protective immune response. Thus, α-glucan-based extract can be used in the prevention of diseases caused by human pathogenic coronaviruses, including SARS-CoV-2 due to its ability to initiate protective immune response (Di, Bertuccioli & Cavecchia, 2020). A proposed mechanism for anti-SARS-CoV-2 action exhibited by polysaccharides has been depicted in Fig. 4.

(Figure reprinted from Xiangyan, Wenwei, Guixiang and Xia (2020), Application prospect of polysaccharides in the development of anti-novel coronavirus drugs and vaccines. International Journal of Biological Macromolecules. 164, 331–343, Copyright (2020) with permission from Elsevier).

### 5.1. Glycan-based molecules as drugs for COVID-19

Glycans are key surface components of the host cells and most viruses and play a crucial role in the viral infections. The presence of glycans on the envelope of viruses such as SARS-CoV-2, HIV, HCV, etc. is found to play an imperative role in virus entry, and the evasion from the immuno-



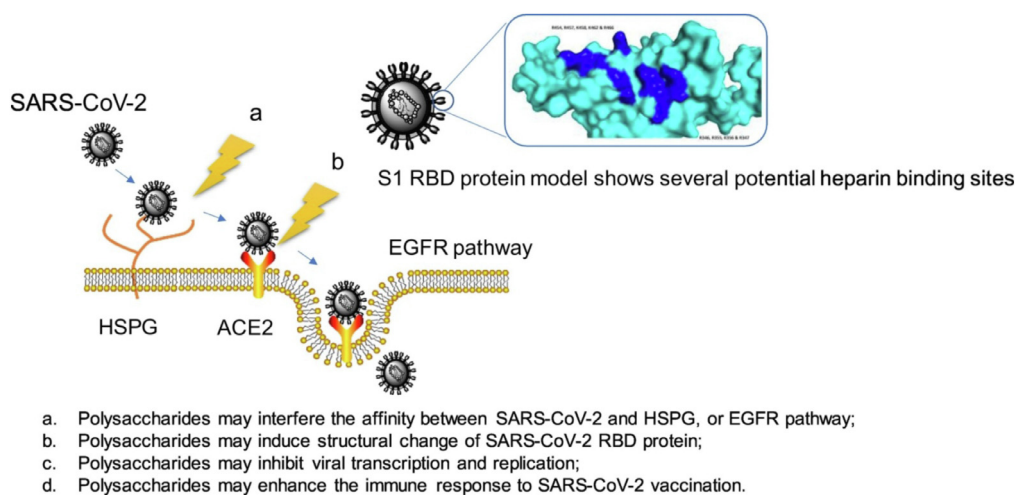


Fig. 4.. The proposed mechanisms of polysaccharides anti-SARS-CoV-2.

logical surveillance of the host. In designing glycan-based therapeutics, the viral protein-glycan interaction is needed to be taken into consideration. For instance, the neuraminidase (NA) of the virus is of vital importance in the replication IAV by hydrolysis of sialic acid (SA) to facilitate the release of virions from infected cells. 2-Deoxy-2,3-didehydro-N-acetylneuraminic acid (DANA), a small molecule analogue of SA that can bind to and block the extremely preserved SA-binding pocket of the enzyme were designed after determining the crystal structure of NA. The DANA was first reported as a NA inhibitor thereby showing antiviral activity against IAV (Shafti-Keramat et al., 2003).

Like other viruses, the envelope of SARS-CoV-2 is also composed of glycoproteins such as S-protein and M-protein (Ahmed, Quadeer & McKay, 2020; Shajahan, Supekar, Gleinich & Azadi, 2020). S-protein has shown to play a significant role in viral pathogenesis and is therefore, a chief target for drug design (Ahmed et al., 2020). In addition, nonstructural (3a) protein observed in glycosylated forms also exhibits a critical role in COVID-19 virulence (Issa, Merhi, Panossian, Salloum & Tokajian, 2020). Carbohydrate-binding agents (CBA) or glycan analogues that interact with the viral-envelope glycans can prevent virus entry into the host cell, thereby preventing the spread of viral infections, opening up new avenues for vaccine development.

Glycan-based molecules such as CS and their sulfated derivatives, CRG and its derivatives, alginate, heparin, dextran, fucans and their derivatives, and green seaweed-derived polysaccharides are reported to show potent antiviral activity against different types of viruses such as CMV, HIV, HCV, HSV, HRV, NDV, African swine fever virus, dengue virus, Japanese encephalitis virus, etc. (Cui et al., 2019; Grice & Mariottini, 2018). For instance, Iota-carrageenan was found to be a potent inhibitor of IAV and it act mainly via interfering virus adsorption to the vaginal flora (McLean et al., 2010). Sulfated polymannuroguluronate (SPMG), a polysaccharide obtained from alginate has served as novel anti-HIV candidate and has been tested in phase II clinic trials in Thailand (Liu et al., 2005). Besides, its anti-HIV mechanism has been completely investigated in China (Xin, Geng, Li, Guan & Li, 2000). Fucans (sulfated glycans) such as fucoidan demonstrated potent antiviral activity against IAV. Moreover, oral administration of fucoidan has resulted in enhanced production of nAbs in the mucosa and blood of IAV infected mice (Hayashi, Lee, Nakano & Hayashi, 2013). Several glycan-based antiviral drug molecules in the clinical trials are depicted in Table 3.

The case studies enlisted in this section clearly suggest that glycan-based (CBA) molecules could be promising candidates in the effective treatment of infections caused by the viruses exhibiting glycoproteins in the envelope including SARS-CoV-2. Moreover, the auxiliary bacterial co-infections are generally present in COVID-19 patients which cause serious health problems. Thus, the glycan-based drug molecules could

also be useful to treat auxiliary bacterial co-infections (MacIntyre et al., 2018) as a result of their antibacterial activity.

## 5.2. Carbohydrate-based drug delivery systems with focus on COVID-19 targeting

Carbohydrates play a crucial role in the identification and mediation of a large number of biological and pathological processes (Gabius, 2009). Carbohydrates (glycoproteins and glycolipids) are presented at the cell surfaces as multivalent domains. The adhesion of pathogens including viruses to the host cell surface occurs through carbohydrate-mediated interactions. Besides, the various glycosylation patterns are also expressed in viruses. Different carbohydrate-based drug delivery systems developed for antiviral drug delivery includes CS NPs, sulfated-CS NPs, alginate NPs, dextran NPs, dextran-functionalized NPs, etc. (Soeun, Yuntao, Peter & Martina, 2019). The carbohydrate-based NPs that mimic the host or pathogen glycoconjugates (glycoproteins, glycolipids, and polysaccharides) have been reported to be a promising approach in the targeting of different types of viral infections. Furthermore, mimicking of glycosylation pattern expressed in the viruses was found to prevent adhesion of virus to host surface. For instance, multivalent gold (Au) NPs decorated with different structural motifs of mannose-type oligosaccharides of gp120 have inhibited the lectin dendritic cell-specific intercellular adhesion molecule-grabbing non-integrin mediated HIV-1 trans-infection of T-cells (Geijtenbeek et al., 2000). In another study, Papp and group have designed SA-functionalized AuNPs which showed the inhibition of influenza virus through receptor targeting (Papp et al., 2010). Moreover, heparan sulfate proteoglycans (HSPG) mimicked AuNPs displayed a potent antiviral activity against diverse viruses such as HSV, HPV, lentivirus, respiratory syncytial virus (RSV), and dengue virus (Cagno et al., 2018). Like all other viruses, the SARS-CoV-2 envelope also exhibits glycoproteins such as S-protein and M-protein. Therefore, carbohydrate-based AuNPs could also be used for effective treatment of COVID-19. Table 4 enlists carbohydrates based drug delivery systems that have been developed against numerous viral infections.

The clinical applications of a wide range of antiviral drugs are limited due to poor solubility, non-specific distribution, enzymatic metabolism, hurdles in drug transportation through the biological membrane, low bioavailability, lack of drug targeting, and associated side effects. To overcome these issues, different types of nano drug delivery systems (DDS) including carbohydrate-based DDS have been developed to achieve drug targeting. NCs such as micelles, nanoparticles, nanogels, etc. prepared using carbohydrate polymers (chitosan, pullulan, etc.) or functionalized with ligands like carbohydrates and can be



**Table 3**

Glycan-based antiviral drug molecules in the clinical trials (Cui et al., 2019)

(Table reproduced from Cui et al. (2019)). Marine glycan-based antiviral agents in clinical or preclinical trials. Reviews in Medical Virology, 29(3), e2043., Copyright (2019) with permission from John Wiley and Sons).

| Glycan-based antiviral molecules | Specific glycans               | Indication    | Clinical trial phases |
|----------------------------------|--------------------------------|---------------|-----------------------|
| Dextran sulfate                  | Dextran                        | HIV           | I & II                |
| SPMG (911)                       | Sulfated-polymannuroguluronate | HIV           | II                    |
| Carraguard                       | $\lambda$ -carrageenan         | HIV, HPV, HSV | II                    |
| Carrageenan nasal spray          | $\iota$ -carrageenan           | IAV           | IV                    |

**Table 4**

Carbohydrate-based drug delivery systems developed against different viruses.

| Disease                   | Antiviral drug                | Type of carbohydrate-based nanodelivery system                           | Key findings  |
|---------------------------|-------------------------------|--|---|
| HCV                       | Curcumin                      | Chitosan NPs   | Remarkable antiviral activities against human hepatoma (Huh7) cells (Loutfy et al., 2020).  |
| Influenza virus infection | SiRNA                         | Chitosan NPs   | Significant uptake of NPs by Vero cells and increased inhibition of influenza virus replication. Increased protection observed in BALB/c mice against lethal influenza virus (Jamali et al., 2018). |
| HIV                       | Zidovudine                    | Dextran hybrid NPs   | Controlled drug release, and improved drug uptake by neuro brain cells and glioma cells (Joshy et al., 2018).   |
| HIV                       | Rilpivirine                   | Cyclodextrin-based nanosponges   | Improved solubility and oral bioavailability (Monica et al., 2018).   |
| HIV                       | Lamivudine                    | Chitosan cross-linked with glutaraldehyde NPs                            | Improved brain targeting (Cheung et al., 2015).   |
| HIV                       | Tenofovir                     | Thiolated-chitosan NPs (TC-NPs) and non-thiolated-chitosan NPs (NTC-NPs) | The TC-NPs exhibited controlled release and significant mucoadhesion compared to NTC-NPs (Jianing, Tao, Vivek, Miezian & Bi-Botti, 2014).   |
| HIV                       | Saquinavir                    | Chitosan NPs   | Improved cell targeting, cellular uptake and antiviral efficacy (Lakshmi, Shilpee, Swaminathan, Udaykumar & Uma, 2014).   |
| HIV                       | Tenofovir disoproxil fumarate | Chitosan microspheres  | Improved mucoadhesion of chitosan microspheres (Arshad & Ram, 2014).  |
| Hepatitis B               | Lamivudine stearate           | Stearic acid-grafted chitosan oligosaccharide polymeric micelles         | Increased loading, increased cellular uptake in HBV-transfected human hepatoblastoma cells, significant <i>in vitro</i> anti-HBV activities (Qian et al., 2010).                                    |

utilized to target specific pathogens (Atanase, Desbrieres & Riess, 2017). Moreover, polysaccharide-based pro-drugs (amphiphilic in nature) can self-assemble into micelles, nanoparticles, and nanogels, thereby improving therapeutic performance of antiviral drugs (Debele, Mekuria & Tsai, 2016; Swierczewska, Han, Kim, Park & Lee, 2016; Zhang, Wardwell & Bader, 2013). The charge, size, shape, surface functionality, and composition of NPs can contribute towards antiviral property and enhance drug targeting and uptake, thus reducing severe side effects. Jamali and co-workers have developed siRNA-loaded CS NPs which effectively targeted nucleoprotein of influenza virus and reduced infection (Jamali et al., 2018). Lamivudine-loaded CS (cross-linked with glutaraldehyde) polymeric NPs developed by Cheung et al., displayed improved brain targeting in HIV (Cheung, Ng, Wong & Chan, 2015). Joshy and group have developed hybrid dextran NPs loaded with zidovudine (ZDV) against HIV/AIDS. The increased cellular internalization of ZDV was observed from NPs when compared to free ZDV (Joshy et al., 2018). In another study, Monica and co-workers have developed rilpivirine (RPV)-loaded cyclodextrin-based nanosponges with the objective to improve the solubility and bioavailability of RPV. The solubility of RPV was found to be increased significantly (10–13 folds) in distilled water in nanosponges. Moreover, the bioavailability studies in Wistar albino rat displayed around 2-fold increase in the oral bioavailability of RPV from nanosponges (Monica, Jagruti, Francesco & Fabrizio, 2018).

As the carbohydrate polymers are also reported to show antiviral activity, the delivery of antiviral drugs using carbohydrate-based NPs can exhibit a synergistic effect. Loutfy et al., have designed curcumin encapsulated chitosan NPs against HCV. The NPs exhibited remarkable anti HCV activity and that was attributed to the synergistic effect of curcumin and chitosan (Loutfy et al., 2020).

To date, no approved drug is specifically available against SARS-CoV-2. Various repurposed drugs such as favipiravir, remdesivir, ritonavir in combination with interferon- $\beta$ , and hydroxychloroquine are widely used for treating COVID-19 infection, however; these drugs are associated with serious side effects (Lai, Shih, Ko, Tang & Hsueh, 2020). These issues can be resolved by delivering them using carbohydrate-based NPs which can improve their solubility, bioavailability, and targeting, and reduce their side effects.

### 5.3. Carbohydrate-based stimuli responsive nanocarriers with focus on COVID-19

Nowadays, the antiviral agents available for treating a variety of pathogenic infections are becoming ineffective because of increased resistance developed by pathogens. Besides, the applications of a number of antiviral drugs are limited due to poor therapeutic performance and associated serious side effects. Stimuli-responsive NCs, compared to non-responsive NCs, have promising applications in the improvement of therapeutic efficacy and reduction of side effects by controlling the release of antiviral drugs precisely to the target site in response to various stimuli (Malobika & Amisha, 2020; Parboosing, Maguire, Govender & Kruger, 2012). Different microenvironmental changes associated with pathogenic infected areas include reduced pH, increased glutathione concentration, enzyme secretion, and allied pathological conditions of infections (Lee, Thompson, 2017; Narang & Mahato, 2010). Polysaccharide based NCs are the most widely used natural NCs because of various noteworthy features of polysaccharides such as high biocompatibility and biodegradability, minimum toxicity, ease of enzymatic or chemical modification, and surface charge that interact with biopharmaceuticals (Boddohi & Kipper, 2010). The differ-

ent stimuli-responsive polysaccharide-based NCs developed and used to treat pathogenic infections include pH-sensitive chitosan and hyaluronic acid (HA) NCs, reduction-sensitive cysteine crosslinked alginate NCs, thermo-sensitive NCs prepared by an amalgamation of polysaccharides with thermo-responsive polymers, and enzyme-sensitive pullulan and HA NCs (Myrick, Venda & Krishnan, 2014). Nadia et al., have developed acyclovir (ACV)-loaded chitosan/xanthan gum-based pH-sensitive topical hydrogels for targeting HSV. The hydrogel has displayed improved absorption and pH-dependent release of ACV (Nadia et al., 2020). In another study, Ting-Ting et al., have developed thermosensitive chitosan hydrogels containing polymeric microspheres for vaginal delivery of tenofovir (TFV) to target HIV. The hydrogel demonstrated a temperature-sensitive release of TFV (Ting-Ting et al., 2017). Furthermore, Shakera and co-workers have developed doxorubicin-loaded hypoxia-responsive mesoporous silica NPs with 4-nitroimidazole- $\beta$ -cyclodextrin. These NPs exhibited higher toxicity to hypoxic cells *in vivo* than normoxic cells (Shakera et al., 2018).

Diverse micro-environmental changes associated with COVID-19 infection include raised body temperature (BT), acidosis, hypoxia and increased level of liver enzymes. The BT of SARS-CoV-2 infected patient was found more than 37 °C (Serena, Koichi, Satoshi & Kiyotake, 2020). The severe metabolic acidosis with multiple organ failure was also observed in COVID-19 infected patients (Shabnam et al., 2020). Besides, these patients also suffered with hypoxia (Serena et al., 2020). Moreover, various case studies reported significant increase in the level of liver alanine aminotransferase (ALT) (Shao-Rui et al., 2020). Thus, SARS-CoV-2 infection is associated with inflammation, hypoxia, oxidative stress, vascular leak, mitochondrial dysfunction, DNA damage, and lung coagulopathy (François et al., 2020).

The different pathological hallmarks of COVID-19 as mentioned above can be utilized in the development of therapeutics targeting SARS-CoV-2 infected lungs and other organs. Further, the pathological conditions favor the development of targeted carbohydrate-based NPs for passive accumulation (via leaky vasculature), active targeted nanoparticles using targeting ligands (carbohydrates) interacting with over expressed epitopes or other cellular components, and internal stimuli (pH, temperature and redox responsive) responsive nanoparticles.

## 6. Concluding remarks and future perspectives

SARS-CoV-2 is an emerging pathogen and is the cause of a pandemic outbreak around the globe. Upon detailed structural (morphological) analysis of the SARS-CoV-2, the S-glycoprotein is found to play a key role in the entry of the virus into the host. The sudden COVID-19 outbreak has sparked worldwide initiative towards the investigation of fast and economic diagnostic technology, effective prophylactic and treatment strategies including novel vaccines and drug candidates. However, the multiple mutations produced in the SARS-CoV-2 have created a massive challenge in the development of these novel diagnostic techniques and treatment strategies. As the surface of different viruses is covered by carbohydrates (glycans), carbohydrate-based diagnostic techniques have been developed to aid in successful diagnosis of the viruses. The S-protein of the SARS-CoV-2 also possesses large number of glycosylation sites which are generally occupied by glycans. Thus, these surface glycans can be targeted for the development of diagnostic techniques for COVID-19 infection. Moreover, as the way of SARS-CoV-2 interaction with its glycan receptor is seasonally consistent, it will be still diagnosed though the genetic code of the virus mutates. Thus, the glycan-based diagnostic technique focused on host-pathogen glycan recognition could become crucial and more universal in the early diagnosis of COVID-19, and could stay effective and sensitive forever.

Development of vaccines is a prophylactic strategy to curtail the transmission of viral diseases including COVID-19. The currently approved vaccines are generally include the use of inactivated viruses, and are mRNA-based vaccines. However, the existence of new variants of the SARS-CoV-2 would make these vaccines less effective. Thus, the glycan-

based vaccines could be a promising alternative approach in the prevention of COVID-19 infections in the future. Besides, the investigation of rationale combination of these glycan-based vaccines is essential in defeating the newly produced variants of the SARS-CoV-2. However, an in-depth understanding of the immune activation pathways of different carbohydrates is very much essential in development of carbohydrate-based safe and effective vaccines.

Although, the glycan-based antiviral drug molecules are in the different phases of clinical trials, sufficient time and extensive research are still needed for their clinical translation. The diverse carbohydrate-based nanoparticles have showed promising potential as carriers for the delivery of vaccines and antiviral drugs with improved targeting, therapeutic performance and reduced side effects against other viruses. Thus, a similar approach can be explored to deliver vaccines and therapeutics with improved therapeutic performance against SARS-CoV-2.

The research related to COVID-19 is continuously evolving by the day and it will be a long wait before we have validated alternatives for sensitive and rapid carbohydrate-based diagnostic techniques, vaccines effective against diverse mutations, and effective glycan-based drug molecules to fight against COVID-19 infection.

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## Declaration of Competing Interest

None.

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