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Mini Review

A new possible indication of direct-acting anti-hepatitis C drugs in the therapeutic management of COVID-19: A narrative literature review

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Abstract

COVID-19 is the latest human crisis to hit most parts of the world since its emergence in China. High prevalence, rapid transmission, and high mortality rates make it necessary to find an effective therapy immediately. Repurposing the available drugs with well-known side effects is proven to be a viable and efficient way to treat this disease. The study purposed to evaluate the therapeutic effect of direct-acting anti-hepatitis C drugs in COVID-19 by reviewing the articles in PubMed, Scopus, Google Scholar, and Embase databases. Some studies mentioned a high similarity between the hepatitis C virus and coronavirus in genome structure and molecular properties. Accordingly, anti-HCV drugs can have a good preventive effect on the actions of SARS-CoV-2 structural and nonstructural proteins. Molecular docking results have shown that anti-hepatitis C drugs such as sofosbuvir, daclatasvir, simeprevir, and elbasvir tend to form various stable bonds with the active sites of essential SARS-CoV-2 proteins. So these drugs can disrupt viral replication and its pathogenesis. Among anti-HCV drugs, sofosbuvir and daclatasvir work efficiently in molecular and human studies. In some human studies, the addition of sofosbuvir/daclatasvir to the therapy of COVID-19 resulted in a shorter duration of hospitalization and higher recovery rates. However, the confirmation of these medications needs more detailed clinical studies in large patient populations.

Keywords: COVID-19, SARS-CoV-2, Coronavirus, Hepatitis C, Antiviral, Sofosbuvir

Introduction

COVID-19 was first confirmed in December 2019, and the causative virus is spreading throughout the world.[1] The severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) as a novel coronavirus created a pandemic in the world.[2] Patients can vary from asymptomatic to mild symptomatic and even to have acute respiratory distress syndrome. In addition to the respiratory system, coronavirus can cause multiorgan dysfunction by infecting other organs such as the intestine, liver, and kidney.[3] Although some drugs have been promising in treating COVID-19, no cure has been approved yet.[4]

Translation of the SARS-CoV-2 genome produces different structural and nonstructural functional proteins. Structural proteins include spike, nucleocapsid, matrix, and envelope

proteins are responsible for the formation of the virus particle. Papain-like protease (PLpro), main protease (Mpro) also named 3-chymotrypsin-like protease (3CLpro), RNA-dependent RNA polymerase (RdRp), and helicase are nonstructural proteins. They play an essential role in viral pathogenesis.[5, 6] RdRp has a critical role in the replication of SARS-CoV-2 RNA and catalyzed new RNA synthesis. Also, helicase is vital in viral replication and proliferation, and Mpro involves producing various functional proteins.[5]

Anti-HCV drugs have been studied in infections caused by the West Nile virus, yellow fever virus (YFV), zika virus (ZIKV), and chikungunya virus, which have similarities with SARS-CoV-2 in genome characteristics. For example, in vitro study by Dragoni et al. in the hepatic and neuronal cell lines determined the antiviral effect of sofosbuvir against the West Nile virus. Besides, the purified West Nile virus RdRp was inhibited dose-dependently by sofosbuvir.[7] Also, Ferreira et al. stated that sofosbuvir inhibited chikungunya virus replication in hepatoma cells (Huh-7) and human astrocyte cells, although it was more effective in Huh-7 than astrocytes.[8] Sofosbuvir interacted with similar conserved NS5-RdRp domains in HCV and YFV. As the authors mentioned, sofosbuvir inhibited the YFV replication in both Huh-7 and Vero cell models. Also, it could decrease the viral load in two patients.[9] Sofosbuvir may induce RdRp function errors. It inhibited the replication of ZIKV in cellular models, including Huh-7 and neuroblastoma cells.[10] Recently, a cell-based study noted that sofosbuvir in the concentration of 100 µM inhibited MERS-CoV RdRp activity by about 40%. Also, dasabuvir in the concentration of 10 µM decreased this RdRp activity by 50% without any cell cytotoxicity.

Both of SARS-CoV-2 and hepatitis C viruses showed similarity in molecular structure and replication mechanism.[11] Therefore, hepatitis C antiviral drugs are hypothesized to be helpful in the treatment of COVID-19. As a brief description, when the hepatitis C virus enters the cell, the RNA of the virus is translated into various proteins using the host system. The NS3/4A protease has an essential role in the post-translational process. NS5B polymerase is necessary for the synthesis of new viruses. Another essential protein is NS5A, which regulates the virus replication and acts in virus assembly. Therefore, the direct-acting anti-HCV drugs are classified into three groups: the first group is NS3/4A protease inhibitors, including glecaprevir, grazoprevir, paritaprevir, boceprevir, sovaprevir, simeprevir, and voxilaprevir. The second group is NS5A protein inhibitors, including daclatasvir, elbasvir, ledipasvir, ombitasvir, pibrentasvir,

and velpatasvir. The third group of drugs is NS5B RdRp inhibitors, which include nucleotide analogs and non-nucleotide analogs. As a nucleotide analog, sofosbuvir has high potency and effectively treats six genotypes of the hepatitis C virus. Dasabuvir, a non-nucleotide analog, has lower potency than other drug classes.[12] Due to the similarities between the hepatitis C virus and SARS-CoV-2, several studies have proposed the use of anti-hepatitis C drugs for further research and clinical trials.

Method

The articles were collected by searching in online databases including PubMed, Scopus, Google Scholar, and Embase. The eligible articles regarding the effects of direct-acting anti-HCV drugs on COVID-19 were selected. We considered the keywords as follows: "Covid" "Corona" "ncov" "COVID-19" "Glecaprevir" "Grazoprevir" "Paritaprevir" "Boceprevir" "Sovaprevir" "Simeprevir" "Voxilaprevir" "Daclatasvir" "Elbasvir" "Ledipasvir" "Ombitasvir" "Pibrentasvir" "Velpatasvir" "Sofosbuvir" "Dasabuvir" "NS3/4A" "NS5A" and "NS5B". Twenty-three most relevant articles were gathered. Accordingly, the molecular docking studies, in vitro studies, and clinical trials were reviewed in this paper. Also, the duplicate publications were excluded. The articles were available online, in English, and without a publication date limitation. The retrieved articles were summarized based on the type of studies in tables 1 and 2.

1. Structural proteins targeting

Spike and nucleocapsid proteins

Simeprevir, Paritaprevir, Glecaprevir, Ledipasvir, Elbasvir, Grazoprevir, Sovaprevir Daclatasvir, Ombitasvir

Studies have shown that angiotensin-converting enzyme 2 (ACE2) receptors are highly expressed on the surface of alveolar epithelial cells and targeted by the coronavirus. The virus detects the ACE2 receptor using the spike (S) protein and can cross the cell membrane barrier and enters the cell. The S protein has two subunits, the S1 subunit interacts with the ACE2 receptor, and the S2 subunit acts to fuse the virus and host cell membranes.[13] Actually, the fourteenth amino acid in the S1 subunit interacts with the ACE2 receptor through a protected

receptor binding domain (RBD). Also, sequence changes at the S protein cleavage site are important for virus pathogenesis.[14]

Simeprevir binds with a high affinity to the S protein of the virus and prevents its interaction with the ACE2 receptor. The drug molecule interacts with the side chains in the RBD and prevents recognition of ACE2 receptor by the S protein. So simeprevir could be considered as a proposed drug for COVID-19 treatment.[15] It has been shown that paritaprevir, glecaprevir, simeprevir, and ledipasvir can bind tightly to this active site with one, two, two, and six hydrogen bonds in the interaction site of the S protein and the receptor, respectively. Also, it was mentioned that paritaprevir and simeprevir have appropriate protein binding energies.[14]

Elbasvir, grazoprevir, and sovaprevir have been identified as potential binders to the RBD structure of S protein. As a result, virus entry to the cell is prevented by interfering with RBD binding to its receptor. Interestingly, sovaprevir forms seven hydrogen bonds with RBD.[16]
Transmembrane protease serine-type 2 (TMPRSS2) is a serine protease that activates the viral S-ACE2 complex as a host protease. Daclatasvir, ombitasvir, and paritaprevir showed high affinity for TMPRSS2 using the sequence-based affinity prediction method that identifies potent target inhibitors with K_d< 100 nM. Daclatasvir, ombitasvir, and paritaprevir appear to have strong inhibitory potency with K_d values less than 10 nM (6.65 nM, 5.91 nM, and 8.75 nM, respectively).[17]

The Mpro and S protein domains are highly stable in SARS-CoV-2. Therefore, it is difficult to establish instability in the integration of these proteins with drugs. According to the Ahmed et al. study, the antiviral activity of simeprevir against SARS-CoV-2 can be increased by using combination therapy with caulerpin and its derivatives.[18]

Nucleocapsid (N) protein is another structural protein that is important in genome replication, transcription, and packaging.[19] An in silico study assessed 548 antiviral agents to identify inhibitor compounds of SARS-CoV-2 structural proteins. In this study, two anti-HCV agents, simeprevir and grazoprevir had a high affinity for binding to N protein. These potential drugs showed a strong tendency to N protein with low binding energy. Simeprevir probably prevent N protein assembly by forming hydrogen bonds with Thr91 and Arg93 in the active site.[1] The in

vitro studies using anti-hepatitis C drugs against essential proteins of SARS-CoV-2 are summarized in table 1.

2. Nonstructural proteins targeting

2.1 RNA dependent RNA polymerase (RdRp)

Sofosbuvir, Ledipasvir, Velpatasvir, Daclatasvir

RdRp is a vital enzyme for RNA transcription and replication. All RNA viruses and some DNA viruses need RdRp for genome replication. The catalytic mechanisms of RdRps are similar in both SARS-CoV-2 and hepatitis C virus.[20] The surface of two aspartate residues in the active site of RdRp is available by a nucleotide channel.[21] When the virus entered the host cell, RdRp is made from its precursors and controls RNA strand length by adding nucleotides to the strand. So, the nucleotide analogs can insert into the RNA chain and stop the RNA elongation.[20]

Sofosbuvir is approved for the treatment of hepatitis C. Also, the drug has excellent results against other viruses like the ZIKV.[22] Sofosbuvir especially can be useful in the early phase of COVID-19 before the virus attacks the lung cells.[23] Recent studies showed that the active form of sofosbuvir terminates RdRp activity.[11, 24] Cellular enzymes convert sofosbuvir to its active triphosphate form. The drug competes with the physiologic ribonucleotide substrates for binding to RdRp.[11, 21] So, modified nucleotide analogs can sit instead of the primary substrates and inhibit RdRp activity. Recent research indicated that 2' position changes in the chemical structure of sofosbuvir prevent nucleotide incorporation by SARS-CoV-2 RdRp and stop RNA strand extension.[11, 25]

The molecular docking process predicted that sofosbuvir might have an inhibitory effect on SARS-CoV-2 RdRp.[24, 26] Elfiky focused on nucleotide inhibitors using homology modeling for building a SARS-CoV-2 RdRp model. This study found that sofosbuvir could establish seven hydrogen bonds and two hydrophobic bonds with the enzyme RdRp. Also, it has metal interaction with magnesium ions in the catalytic residue, which increases the drug stability at the site of action.[21] In another study, Elfiky mentioned that sofosbuvir had comparable binding energies to the native nucleotides.[27] Also, Chien et al. study demonstrated that sofosbuvir entered into an RNA primer via SARS-CoV-2 RdRp and terminated the polymerase interaction.[24]

Sofosbuvir has a well-known safety profile; however, studies have shown that the virus is more likely to become resistant to this drug. Therefore, appropriate strategies should be considered to prevent this issue.[28] In Chen et al. study, a model for Mpro was designed based on the crystal structure. According to this molecular model, they proposed 16 drug candidates for further research. They found that ledipasvir or velpatasvir with low side effects combined with sofosbuvir could have an inhibitory effect on SARS-CoV-2. They stopped virus growth through two mechanisms that reduced the possibility of drug resistance.[29, 30]

Few clinical trials used direct-acting anti-HCV drugs in the treatment of COVID-19. In a clinical study, two patient groups received sofosbuvir/daclatasvir or ribavirin in addition to the standard treatment (single dose hydroxychloroquine + lopinavir/ritonavir). Sofosbuvir/daclatasvir group had a significantly lower mortality rate than the ribavirin group (5.7% and 33%, respectively) ($p= 0.01$). Also, the duration of hospital stay in patients who received sofosbuvir/daclatasvir was 5 days, whereas it was 9 days in the ribavirin group ($p< 0.01$). This open-label study was revealed that the treatment with sofosbuvir/daclatasvir was more effective and had lower side effects than ribavirin. Although the study process showed that the patients were well distributed in the groups, a complete randomized and blind study was not performed. Also, It was better to have a control group that does not receive ribavirin for comparison between groups.[31]

Another open-label research was performed on 66 patients who suffered from moderate to severe COVID-19. Patients in the intervention group received sofosbuvir and daclatasvir plus standard therapy, including hydroxychloroquine with or without lopinavir/ritonavir. Patients in the control group received only standard care. 88% and 67% of patients in the intervention and control groups achieved recovery within 14 days ($p= 0.076$), respectively. The median duration of hospitalization was 6 and 8 days in the intervention and control groups ($p= 0.029$), respectively. Also, the number of patients discharged from the hospital was significantly higher in the treatment group than in the control group ($p= 0.041$). However, the lower mortality rate in the intervention group than the control group was not significant. As a limitation of the study, fewer patients received lopinavir/ritonavir in the intervention arm (33% vs. 64% in the control group, $p= 0.026$). Besides, the small patient population was another limitation of the study.[32]

A randomized clinical trial was accomplished in patients with moderate severity COVID-19. The intervention group received combination therapy with sofosbuvir, daclatasvir, and ribavirin, while the control group received standard treatment (hydroxychloroquine + lopinavir/ritonavir ± ribavirin). In this study, no significant differences in mortality and ICU admission rates were reported between the groups. However, the incidence of recovery was significantly higher in the intervention group (P= 0.033). A significantly higher number of diabetic patients in the control group was a limitation of the study. Also, the study population was not large enough to confirm the beneficial effects of sofosbuvir/daclatasvir on survival.[33] The human studies on the effects of anti-HCV drugs in COVID-19 are summarized in table 2.

2.2 Protease

Elbasvir, Grazoprevir, Glecaprevir, Paritaprevir, Simeprevir, Ledipasvir, Raltegravir, Ombitasvir, Daclatasvir, Boceprevir

After the virus enters the host cell and essential viral proteins are expressed, two enzymes, namely Mpro and PLpro, break down the polyproteins into smaller products used in virus replication and transcription.[13] High sequence conservation and vital role of Mpro in the pathogenesis of COVID-19 resulted in targeting Mpro for designing and developing structure-based drugs. Recent studies revealed that HCV NS5B polymerase interacts with the His41, Cys145, His57, and Ser139 catalytic dyads in the Mpro. Similarities of proteases in both SARS-CoV-2 and HCV make the HCV NS3/4A protease inhibitors, probable drug candidates to block the SARS-CoV-2 Mpro transcription function.[4]

Although there is low sequence identity among SARS-CoV-2 and hepatitis C virus Mpro, the co-crystal ligands greatly overlap. So, repurposing anti-HCV drugs may efficiently work against SARS-CoV-2. Accordingly, the docking process determined that elbasvir has inhibitory activities against SARS-CoV-2 Mpro.[34] Another FDA-approved drug against the hepatitis C virus is grazoprevir which has stronger binding potential to SARS-CoV-2 Mpro compared with binding to HCV NS3/4A.[5]

Glecaprevir is a drug approved against the hepatitis C virus with NS3/4A protease inhibitory mechanism. The drug has a high affinity to the binding site of SARS-CoV-2 Mpro. Glecaprevir binds tightly to Mpro with various hydrogen bonds and hydrophobic interactions and can stop the virus replication.[35] Virtual screening showed that glecaprevir binds to the active site of Mpro and form non-covalent interactions with conserved residues of the Mpro binding pocket. So, glecaprevir can be promising for the therapeutic management of COVID-19.[36]

It was demonstrated that simeprevir and grazoprevir could form hydrogen bonds with protease binding pocket. One study reported that simeprevir could have four hydrogen bonds at this active site, of which three interactions were constant. This study indicated that simeprevir was stable in the protease binding pocket and had suitable interaction with catalytic residues Cys145 and His41.[37] According to a molecular docking study conducted by Rahman et al., simeprevir was introduced as one of the best candidates against SARS-CoV-2 Mpro. Simeprevir had several bonds with amino acid residues in addition to hydrogen bonds and electrostatic interactions. The scores plot indicated that the apo–Mpro (protein without ligand) and simeprevir–Mpro complex were the same in energy, structural profile, and structural compactness.[6]

The database screening demonstrated two potential drugs, paritaprevir and simeprevir, as protease inhibitors, which had strong hydrogen bonds with low binding energy scores. Both drug molecules were bound near the catalytic dyad.[38] Paritaprevir and raltegravir can form sufficient interaction with the active site by forming hydrogen bonds with Mpro. Drug-protein complexes were dramatically stable after the drug linked to the active site. Also, the drugs could bind efficiently to the catalytic dyad.[39]

Another in silico study screened 75 FDA-approved drugs against Mpro. These compounds, such as ombitasvir, elbasvir, simeprevir, paritaprevir, daclatasvir, and glecaprevir, had stable interactions with the enzyme by hydrogen bonds and hydrophobic interactions.[2] Also, the analyses of raltegravir, simeprevir, and daclatasvir in reacting process with RdRp and Mpro proteins indicated that these drugs can form a wide variety range of non-covalent bonds with these enzymes and may have promising results in in vitro and in vivo studies.[40]

Conclusion

There is an urgent global need to treat COVID-19 infection. Therefore, we reviewed studies about the use of hepatitis C antiviral drugs to treat this infection. SARS-CoV-2 is similar to the hepatitis C virus in terms of genome structure and molecular properties. We assumed that hepatitis C antivirals could be beneficial in the management of COVID-19 since the previous reports mentioned their effectiveness in other similar viruses like ZIKV, chikungunya, and YFV.

Studies based on computer strategies and molecular modeling, have shown that anti-hepatitis C drugs can be effective against viral proteins and terminate SARS-CoV-2 life cycles. Docking studies measured the binding affinity of ligand-protein and the stability of drugs that target various pivotal proteins like S, N, RdRp, and protease during the simulation. Accordingly, most of the anti-HCV drugs had strong potency for binding to the targets. Also, they were stable in the active site of proteins by forming a variety number of bonds. Paritaprevir, glecaprevir, simeprevir, ledipasvir, and daclatasvir interfere with the virus entry by targeting S protein. Daclatasvir, simeprevir, and grazoprevir showed a strong tendency to N protein, which has a role in RNA packaging. Sofosbuvir can compete with the natural substrate for binding to RdRp which causes mutations in the virus RNA.

Few clinical studies in small patient populations showed that sofosbuvir/daclatasvir reduced hospital length of stay and increased the recovery rate. However, other direct-acting anti-hepatitis C drugs have not been used in clinical trials. Therefore, more human studies with larger sample sizes are needed to validate the therapeutic effects of these candidate drugs.

Authors' contributions

All authors contributed to the design and conception of this review. Mashhadi Hemmatabadi and Jafari prepared the first draft. Hosseinjani performed the literature search and critically revised the draft. All authors approved the final version.

References

1. Bhowmik D, Nandi R, Jagadeesan R, Kumar N, Prakash A, Kumar D. Identification of potential inhibitors against SARS-CoV-2 by targeting proteins responsible for envelope formation and virion assembly using docking based virtual screening, and pharmacokinetics approaches. Infect Genet Evol. 2020;84:104451. doi:10.1016/j.meegid.2020.104451
2. Gurung AB, Ali MA, Lee J, Farah MA, Al-Anazi KM. Structure-based virtual screening of phytochemicals and repurposing of FDA approved antiviral drugs unravels lead molecules as potential inhibitors of coronavirus 3C-like protease enzyme. J King Saud Univ Sci. 2020;32(6):2845-2853. doi:10.1016/j.jksus.2020.07.007
3. Robba C, Battaglini D, Pelosi P, Rocco PR. Multiple organ dysfunction in SARS-CoV-2: MODS-CoV-2. Expert Rev Respir Med. 2020;14(9):865-868. doi: 10.1080/17476348.2020.1778470
4. Havranek B, Islam SM. An in silico approach for identification of novel inhibitors as potential therapeutics targeting COVID-19 main protease. J Biomol Struct Dyn. 2020;1-12. doi:10.1080/07391102.2020.1776158
5. Iftikhar H, Ali HN, Farooq S, Naveed H, Shahzad-ul-Hussan S. Identification of potential inhibitors of three key enzymes of SARS-CoV2 using computational approach. Comput Biol Med. 2020;122:103848. doi:10.1016/j.compbiomed.2020.103848
6. Rahman MM, Saha T, Islam KJ, Suman RH, Biswas S, Rahat EU, et al. Virtual screening, molecular dynamics and structure–activity relationship studies to identify potent approved drugs for Covid-19 treatment. J Biomol Struct Dyn. 2020;1-11. doi:10.1080/07391102.2020.1794974
7. Dragoni F, Boccuto A, Picarazzi F, Giannini A, Giammarino F, Saladini F, et al. Evaluation of sofosbuvir activity and resistance profile against West Nile virus in vitro. Antiviral Res 2020;175:104708. doi:10.1016/j.antiviral.2020.104708
8. Ferreira AC, Reis PA, de Freitas CS, Sacramento CQ, Hoelz LVB, Bastos MM, et al. Beyond members of the Flaviviridae family, sofosbuvir also inhibits chikungunya virus replication. Antimicrob Agents Chemother. 2019;63(2):e01389-18. doi:10.1128/AAC.01389-18

9. Mendes ÉA, de Pilger DRB, Nastri ACdSS, de Mello Malta F, dos Santos Pascoalino B, D'Albuquerque LAC, et al. Sofosbuvir inhibits yellow fever virus in vitro and in patients with acute liver failure. Ann Hepatol. 2019;18(6):816-24. doi:10.1016/j.aohep.2019.09.001
10. Sacramento CQ, De Melo GR, De Freitas CS, Rocha N, Hoelz LVB, Miranda M, et al. The clinically approved antiviral drug sofosbuvir inhibits Zika virus replication. Sci Rep. 2017;7:40920. doi:10.1038/srep40920
11. Ju J, Li X, Kumar S, Jockusch S, Chien M, Tao C, et al. Nucleotide analogues as inhibitors of SARS-CoV polymerase. Pharmacol Res Perspect. 2020;8(6):e00674. doi:10.1002/prp2.674
12. Direct-acting antivirals for the treatment of hepatitis C virus infection. http://www uptodate com/. Accessed 02 February 2017.
13. Huang J, Song W, Huang H, Sun Q. Pharmacological therapeutics targeting RNA-dependent RNA polymerase, proteinase and spike protein: from mechanistic studies to clinical trials for COVID-19. J Clin Med. 2020;9(4):1131. doi:10.3390/jcm9041131
14. Mahdian S, Ebrahim-Habibi A, Zarrabi M. Drug repurposing using computational methods to identify therapeutic options for COVID-19. J Diabetes Metab Disord. 2020;19(2):1-9. doi:10.1007/s40200-020-00546-9
15. Trezza A, Iovinelli D, Santucci A, Prischi F, Spiga O. An integrated drug repurposing strategy for the rapid identification of potential SARS-CoV-2 viral inhibitors. Sci Rep. 2020;10(1):13866. doi:10.1038/s41598-020-70863-9
16. Behloul N, Baha S, Guo Y, Yang Z, Shi R, Meng J. In silico identification of strong binders of the SARS-CoV-2 receptor-binding domain. Eur J Pharmacol. 2020;890:173701. doi:10.1016/j.ejphar.2020.173701
17. Choi Y, Shin B, Kang K, Park S, Beck BR. Target-Centered Drug Repurposing Predictions of Human Angiotensin-Converting Enzyme 2 (ACE2) and Transmembrane Protease Serine Subtype 2 (TMPRSS2) Interacting Approved Drugs for Coronavirus Disease 2019 (COVID-19) Treatment through a Drug-Target Interaction Deep Learning Model. Viruses. 2020;12(11):1325. doi:10.3390/v12111325
18. Ahmed SA, Abdelrheem DA, Abd El-Mageed H, Mohamed HS, Rahman AA, Elsayed KN, et al. Destabilizing the structural integrity of COVID-19 by caulerpin and its derivatives

along with some antiviral drugs: An in silico approaches for a combination therapy. Struct Chem. 2020;31(6):2391-412. doi:10.1007/s11224-020-01586-w

19. Tilocca B, Soggiu A, Sanguinetti M, Musella V, Britti D, Bonizzi L, et al. Comparative computational analysis of SARS-CoV-2 nucleocapsid protein epitopes in taxonomically related coronaviruses. Microbes Infect. 2020;22(4-5):188-194. doi:10.1016/j.micinf.2020.04.002
20. Zhu W, Chen CZ, Gorshkov K, Xu M, Lo DC, Zheng W. RNA-dependent RNA polymerase as a target for COVID-19 drug discovery. SLAS Discov. 2020;25(10):1141-1151. doi:10.1177/2472555220942123
21. Elfiky AA. Anti-HCV, nucleotide inhibitors, repurposing against COVID-19. Life Sci. 2020;248:117477. doi:10.1016/j.lfs.2020.117477
22. Nascimento IJdS, de Aquino TM, da Silva-Júnior EF. Drug Repurposing: A Strategy for Discovering Inhibitors against Emerging Viral Infections. Curr Med Chem. 2020. doi:10.2174/0929867327666200812215852
23. Nourian A, Khalili H. Sofosbuvir as a potential option for the treatment of COVID-19. Acta Biomed. 2020;91(2):236-8. doi:10.23750/abm.v91i2.9609.
24. Chien M, Anderson TK, Jockusch S, Tao C, Kumar S, Li X, et al. Nucleotide Analogues as Inhibitors of SARS-CoV-2 Polymerase, a Key Drug Target for COVID-19. J Proteome Res. 2020;19(11):4690-4697. doi:10.1021/acs.jproteome.0c00392
25. Gordon CJ, Tchesnokov EP, Woolner E, Perry JK, Feng JY, Porter DP, et al. Remdesivir is a direct-acting antiviral that inhibits RNA-dependent RNA polymerase from severe acute respiratory syndrome coronavirus 2 with high potency. J Biol Chem. 2020;295(20):6785-97. doi:10.1074/jbc.RA120.013679
26. Elfiky AA. SARS-CoV-2 RNA dependent RNA polymerase (RdRp) targeting: An in silico perspective. J Biomol Struct Dyn. 2020;1-9. doi:10.1080/07391102.2020.1761882
27. Elfiky AA. Ribavirin, Remdesivir, Sofosbuvir, Galidesivir, and Tenofovir against SARS-CoV-2 RNA dependent RNA polymerase (RdRp): A In silico study. Life Sci. 2020;253:117592. doi:10.1016/j.lfs.2020.117592

28. Jácome R, Campillo-Balderas JA, de León SP, Becerra A, Lazcano A. Sofosbuvir as a potential alternative to treat the SARS-CoV-2 epidemic. Sci Rep. 2020;10(1):9294. doi:10.1038/s41598-020-66440-9
29. Chen YW, Yiu C-PB, Wong K-Y. Prediction of the SARS-CoV-2 (2019-nCoV) 3C-like protease (3CL pro) structure: virtual screening reveals velpatasvir, ledipasvir, and other drug repurposing candidates. F1000Res. 2020;9:129. doi:10.12688/f1000research.22457.2
30. Izzi A, Messina V, Rinaldi L, Maggi P. Editorial–Sofosbuvir/Velpatasvir as a combination with strong potential activity against SARS-CoV2 (COVID-19) infection: how to use direct-acting antivirals as broad-spectrum antiviral agents. Eur Rev Med Pharmacol Sci. 2020; 24(10):5193-4. doi:10.26355/eurrev_202005_21297
31. Eslami G, Mousaviasl S, Radmanesh E, Jelvay S, Bitaraf S, Simmons B, et al. The impact of sofosbuvir/daclatasvir or ribavirin in patients with severe COVID-19. J Antimicrob Chemother. 2020;75(11):3366-3372. doi:10.1093/jac/dkaa331
32. Sadeghi A, Asgari AA, Norouzi A, Kheiri Z, Anushirvani A, Montazeri M, et al. Sofosbuvir and daclatasvir compared with standard of care in the treatment of patients admitted to hospital with moderate or severe coronavirus infection (COVID-19): a randomized controlled trial. J Antimicrob Chemother. 2020;75(11):3379-3385. doi:10.1093/jac/dkaa334
33. Kasgari HA, Moradi S, Shabani AM, Babamahmoodi F, Badabi ARD, Davoudi L, et al. Evaluation of the efficacy of sofosbuvir plus daclatasvir in combination with ribavirin for hospitalized COVID-19 patients with moderate disease compared with standard care: a single-centre, randomized controlled trial. J Antimicrob Chemother. 2020;75(11):3373-3378. doi:10.1093/jac/dkaa332
34. Wang J. Fast identification of possible drug treatment of coronavirus disease-19 (COVID-19) through computational drug repurposing study. J Chem Inf Model. 2020;60(6):3277-3286. doi:10.1021/acs.jcim.0c00179
35. Chtita S, Belhassan A, Aouidate A, Belaidi S, Bouachrine M, Lakhlifi T. Discovery of Potent SARS-CoV-2 Inhibitors from Approved Antiviral Drugs via Docking and Virtual Screening. Comb Chem High Throughput Screen. 2020. doi:10.2174/1386207323999200730205447

36. Shamsi A, Mohammad T, Anwar S, AlAjmi MF, Hussain A, Rehman M, et al. Glecaprevir and Maraviroc are high-affinity inhibitors of SARS-CoV-2 main protease: possible implication in COVID-19 therapy. Biosci Rep. 2020;40(6):BSR20201256. doi:10.1042/BSR20201256
37. Hosseini FS, Amanlou M. Anti-HCV and anti-malaria agent, potential candidates to repurpose for coronavirus infection: Virtual screening, In silico, and molecular dynamics simulation study. Life Sci. 2020;258:118205. doi:10.1016/j.lfs.2020.118205
38. Alamri MA, Tahir Ul Qamar M, Mirza MU, Bhadane R, Alqahtani SM, Muneer I, et al. Pharmacoinformatics and molecular dynamics simulation studies reveal potential covalent and FDA-approved inhibitors of SARS-CoV-2 main protease 3CLpro. J Biomol Struct Dyn. 2020;1-13. doi:10.1080/07391102.2020.1782768
39. Khan RJ, Jha RK, Amera GM, Jain M, Singh E, Pathak A, et al. Targeting SARS-CoV-2: a systematic drug repurposing approach to identify promising inhibitors against 3C-like proteinase and 2′-O-ribose methyltransferase. J Biomol Struct Dyn. 2020;1-14. doi:10.1080/07391102.2020.1753577
40. Ahmed S, Mahtarin R, Ahmed SS, Akter S, Islam MS, Mamun AA, et al. Investigating the binding affinity, interaction, and structure-activity-relationship of 76 prescription antiviral drugs targeting RdRp and Mpro of SARS-CoV-2. J Biomol Struct Dyn. 2020;1-16. doi:10.1080/07391102.2020.1796804
41. Manikyam HK, Joshi SK. Whole Genome Analysis and Targeted Drug Discovery Using Computational Methods and High Throughput Screening Tools for Emerged Novel Coronavirus (2019-nCoV). J Pharm Drug Res. 2020;3(2):341-36

Table 1: Summary of in vitro studies using direct-acting anti-hepatitis C drugs against essential proteins of SARS-CoV-2

Target	Anti-HCV drugs	Study design	Outcome	References
S protein	Paritaprevir Simeprevir Ledipasvir Glecaprevir	2471 FDA-approved drugs against cleavage sites in S protein were screened.	Paritaprevir, glecaprevir, simeprevir, and ledipasvir formed one, two, two, and six hydrogen bonds with S protein. Paritaprevir and simeprevir showed appropriate binding energies to target proteins.	14
S protein	Simeprevir	1582 FDA-approved drugs against S glycoprotein were virtually screened.	Simeprevir bound to the S protein receptor with high affinity and inhibited ACE2 binding. Simeprevir formed several hydrogen and Van Der Waals bonds with amino acid residues.	15
S protein	Elbasvir Grazoprevir Sovaprevir	Molecules were identified based on binding to the RBD residues.	Elbasvir, grazoprevir, and sovaprevir interfered with the formation of the RBD-ACE-2 complex by creating hydrogen bonds with RBD residue, especially for sovaprevir.	16
S protein	Daclatasvir Ombitasvir Paritaprevir	1400 selected drugs from the DrugBank database for binding affinity prediction to ACE2 and TMPRSS2 were Screened.	Daclatasvir, ombitasvir, and paritaprevir appeared to have strong inhibitory potency with K_d values 6.65 nM, 5.91 nM, and 8.75 nM, respectively.	17
N protein	Grazoprevir Simeprevir	Drug candidates among 548 compounds that target structural proteins in SARS-CoV-2 were identified. Binding affinity and the stability of selected agents in the active site were analyzed.	Simeprevir and Grazoprevir were stable in the active site of proteins during the molecular docking simulation. Simeprevir and Grazoprevir showed appropriate docking results for N protein. Simeprevir formed hydrogen bonds with the active site residues.	1
RdRp	Sofosbuvir	RdRp modeling by sequence analysis and docking method was done. Anti-polymerase drugs, sofosbuvir, and ribavirin	Sofosbuvir could form seven hydrogen and two hydrophobic bonds. It also interacted with Mg^{+2} in the catalytic dyad of RdRp.	21

		targeted this model.		
RdRp	Sofosbuvir	SARS-CoV-2 RdRp was modeled and validated. Anti-polymerase drugs targeted this protein.	Sofosbuvir could bind to SARS-CoV-2 RdRp with comparable binding energy to native nucleotides (-7.5 kcal/mol). Sofosbuvir formed seven hydrogen and two hydrophobic bonds with the SARS-CoV-2 RdRp.	27
RdRp	Sofosbuvir	Polymerase extension experiment to identify the effect of sofosbuvir on further incorporation by RdRp was done.	Incorporated Sofosbuvir by SARS-CoV-2 RdRp blocked viral RNA synthesis.	24
Mpro	Glecaprevir Daclatasvir Paritaprevir Ombitasvir Elbasvir Simeprevir	About 263 phytochemicals and 75 antiviral drugs were selected and screened against Mpro.	Glecaprevir had stable hydrogen and hydrophobic interactions with the enzyme. Reported binding free energy: simeprevir= -9.7 kcal/mol, ledipasvir= -9.3 kcal/mol, paritaprevir= -9.3 kcal/mol, glecaprevir= -9.3 kcal/mol, and Daclatasvir= -9.2 kcal/mol	2
Mpro	Glecaprevir	The binding affinity of 2388 FDA-approved drugs to the SARS-CoV-2 Mpro was assayed.	Glecaprevir fitted firmly to the binding pocket of SARS-CoV-2 Mpro. Glecaprevir formed interactions including three hydrogen bonds, and two fluorine interactions.	36
Mpro	Glecaprevir	129 FDA-approved medications, especially antiviral drugs, for targeting the active site of the SARS-CoV-2 Mpro were screened.	Glecaprevir formed hydrogen and hydrophobic bonds with amino acid residues in the active site of the protease. Glecaprevir bound tightly to the Mpro (docking score −9.40 Kcal/mol)	35
Mpro	Grazoprevir	A library of 4574 compounds was screened to find molecules that could bind to essential proteins (RdRp, Mpro, and helicase).	Grazoprevir could potentially interfere with SARS-CoV-2 protease dimer formation. The predicted binding score (-10.1 Kcal/mol) showed the potential stronger binding of grazoprevir to SARS-CoV-2 Mpro than HCV NS3/4A.	5
Mpro	Simeprevir	1615 FDA-approved and 4266 world-approved drugs were screened for interaction with SARS-CoV-2 Mpro.	Simeprevir showed interaction with Mpro by forming three hydrogen bonds with Asn119, His163, and Thr26 and some other hydrophobic bonds. Simeprevir bound to Mpro with a low binding energy	37

			of −11.33 Kcal/mol because of its long and flexible structure.	
Mpro	Simeprevir	1615 FDA-approved drugs against Mpro were screened.	Simeprevir formed various interactions with the Mpro binding site residues. The simeprevir–Mpro complex presented higher binding free energy than the remdesivir–Mpro complex. Also, simeprevir-Mpro was stable over the simulation period.	6
Mpro	Paritaprevir Simeprevir	116 FDA-approved agents were virtually screened for SARS-CoV-2 Mpro inhibition.	Paritaprevir and simeprevir formed hydrogen bonds with amino acid residues near catalytic dyad. Paritaprevir and simeprevir showed low binding energies of -8.8 and -8.78 kcal/mol, respectively.	38
Mpro	Paritaprevir Raltegravir	123 antiviral drugs that target Mpro and 2′-O-ribose methyltransferase were screened.	Both paritaprevir and raltegravir could interact with amino acids Cys145 and His41 in the active site. They also interacted with the substrate-binding pocket. Paritaprevir and raltegravir formed six and five intermolecular hydrogen bonds with Mpro, respectively.	39
Mpro	Elbasvir	Virtual screening was done for 2201 approved drugs targeting Mpro.	Elbasvir had inhibitory effects on SARS-CoV-2 Mpro.	34
RdRp Mpro	Velpatasvir Sofosbuvir Ledipasvir	A three-dimensional model of SARS-CoV-2 Mpro for 16 drug candidates was prepared.	They were very effective because of their inhibitory actions on Mpro and RdRp.	29
RdRp Mpro	Raltegravir Simeprevir Daclatasvir	Binding affinity and interaction of drugs to RdRp and Mpro were assessed.	These drugs formed a greater number of non-covalent bonds with RdRp and Mpro than other antiviral agents and were stable in the target site.	40
RdRp Mpro PLpro	Paritaprevir Sofosbuvir	Paritaprevir, sofosbuvir, and some other drugs were used as ligands for viral proteins such as S, Mpro, PLpro, and RdRp.	Paritaprevir showed docking scores of -7.09 kcal/mol for PLpro and -9.23 kcal/mol for Mpro. Sofosbuvir showed a docking score of -5.40 kcal/mol for RdRp. The results indicated the strong tendency of drugs to the target proteins.	41

HCV= hepatitis C virus, S protein= spike protein, N protein= nucleocapsid protein, ACE2= angiotensin-converting enzyme 2, RBD= receptor-binding domain, RdRp= RNA dependent RNA polymerase, Mpro= main protease,

TMPRSS2= transmembrane protease serine-type 2, SARS-CoV-2= severe acute respiratory syndrome coronavirus 2, PLpro= papain-like protease

Table 2: Summary of human studies using direct-acting anti-hepatitis C drugs in the management of COVID-19

Anti-HCV drugs	Study groups	Outcome	references
Sofosbuvir/ Daclatasvir	The patients (n= 62) were allocated into two groups: First group (sofosbuvir/daclatasvir) (n= 35) Second group (ribavirin) (n= 27)	The mortality rate (5.7% vs. 33%, p= 0.01), mean hospitalization duration (5 days vs. 9 days, p< 0.01), median time to hospital discharge (6 days vs. 11 days, p< 0.01), and ICU admission rate (17% vs. 48%, p= 0.01) were significant lower in the sofosbuvir/daclatasvir arm than in ribavirin arm. Survival probability was significantly higher (log rank p< 0.01) in the sofosbuvir/daclatasvir arm. No significant difference was observed between sofosbuvir/daclatasvir and ribavirin groups in the median duration of ICU stay (3.5 days and 5 days, respectively) (p= 0.24).	31
Sofosbuvir/ Daclatasvir	The patients (n= 66) were divided into two groups: Intervention group (sofosbuvir/daclatasvir) (n = 33) Control group (n= 33)	Significantly higher clinical recovery was observed in the intervention group patients (88% vs. %67) (p= 0.076). The number of deaths in the intervention and control groups was 3 and 5, respectively (p= 0.708). Median duration of hospitalization in the intervention arm was significantly shorter than the control arm (6 days vs. 8 days) (p= 0.029). The number of patients who required invasive mechanical ventilation in the intervention and control groups was 3 and 7, respectively (p= 0.303).	32
Sofosbuvir/ Daclatasvir	The patients (n= 48) were allocated into two groups: Intervention group (sofosbuvir/daclatasvir/ribavirin) (n= 24) Control group (n= 24)	The median duration of hospitalization was the same in both groups (6 days) (p= 0.398). No significant differences in mortality rate (p= 0.234) and ICU admission rate (p= 0.109) were observed between groups. Intervention group showed higher cumulative incidence of recovery (p= 0.033).	33

COVID-19= coronavirus disease 2019, ICU= intensive care unit