Targeting SARS-CoV-2 spike protein of COVID-19 with naturally occurring Phytochemicals: an *In silco* study for drug development

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Abbreviations: COVID-19, Coronavirus Disease 2019, SARS-CoV-2S**,** Severe Acute Respiratory Syndrome Coronavirus 2 Spike Protein, HCQ, Hydroxychloroquine, CQ, Chloroquine, ACE2, Angiotensin Converting Enzyme-2, MERS-CoV, Middle East Respiratory Syndrome coronavirus, PDB, protein data bank, ADME, absorption, distribution, metabolism and excretion

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Abstract

Spike glycoprotein found on the surface of SARS-CoV-2 (SARS-CoV-2S) is a class I fusion protein which helps the virus in its initial attachment with human Angiotensin converting enzyme 2 (ACE2) receptor and its consecutive fusion with the host cells. The attachment is mediated by the S1 subunit of the protein via its receptor binding domain. Upon binding with the receptor the protein changes its conformation from a pre-fusion to a post-fusion form. The membrane fusion and internalization of the virus is brought about by the S2 domain of the spike protein. From ancient times people have relied on naturally occurring substances like phytochemicals to fight against diseases and infection. Among these phytochemicals, flavonoids and non-flavonoids have been found to be the active source of different anti-microbial agents. Recently, studies have shown that these phytochemicals have essential anti-viral activities. We performed a molecular docking study using 10 potential naturally occurring flavonoids/non-flavonoids against the SARS-CoV-2 spike protein and compared their affinity with the FDA approved drug hydroxychloroquine (HCQ). Interestingly, the docking analysis suggested that C-terminal of S1 domain and S2 domain of the spike protein are important for binding with these compounds. Kamferol, curcumin, pterostilbene, and HCQ interact with the C-terminal of S1 domain with binding energies of -7.4, -7.1, -6.7 and -5.6 Kcal/mol, respectively. Fisetin, quercetin, isorhamnetin, genistein, luteolin, resveratrol and apigenin on the other hand, interact with the S2 domain of spike protein with the binding energies of -8.5, -8.5, -8.3, -8.2, -8.2, -7.9, -7.7 Kcal/mol, respectively. Our study suggested that, these flavonoid and non-flavonoid moieties have significantly high binding affinity for the two main important domains of the spike protein which is responsible for the attachment and internalization of the virus in the host cell and their binding affinities are much higher compared to that of HCQ. In addition, ADME (absorption, distribution, metabolism and excretion) analysis also suggested that these compounds consist of drug likeness property which may help for further explore as anti-SARS-CoV-2 agents. Further, *in vitro* and *in vivo* study of these compounds will provide a clear path for the development of novel compounds that would most likely prevent the receptor binding or internalization of the SARS-CoV-2 spike protein and therefore could be used as drugs for COVID-19 therapy.

Introduction: The world population is currently facing a severe challenge for survival due to the rise of a global pandemic named Coronavirus Disease 2019 (COVID-19) [1]. This pandemic is caused by a positive sense RNA virus belonging to the β-coronaviridae genera called severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) [2, 3]. As per the reports provided by the World Health Organization (WHO) currently there exists no effective treatment regime including antivirals or vaccines against SARS-CoV-2. Infected cases along with mortality are increasing rapidly day by day and scientists all over the world are desperately looking for effective compounds which can be used as antivirals to use against this virus. Natural compounds with high bioavailability and low cytotoxicity seem to be the most efficient candidates in this regard. Since ancient times, humans have resorted to the use of natural compounds especially phytochemicals for the treatment of different diseases and disorders [4]. Flavonoids are secondary metabolites produced by plants and play very important roles in plant physiology, possessing a variety of potential biological benefits such as antioxidant, anti-inflammatory, anticancer, antibacterial, antifungal and antiviral activities [5, 6]. Different flavonoids including both flavones and flavonols have been quite thoroughly investigated for their potential antiviral properties and many of them showed significant antiviral response in both *in vitro* as well as *in vivo* studies [7]. Curcumin, a component of turmeric, has been described to exhibit enhanced antiviral activity against diverse viruses such as dengue virus (serotype 2) herpes simplex virus, human immunodeficiency virus, Zika and chikungunya viruses among others [8]. Apigenin isolated from the sweet basil (*Ocimum basilicum*) plant has shown potent antiviral activity against hepatitis B virus, adenoviruses, african swine fever virus and some RNA viruses *in vitro* [9]. Luteolin, another acclaimed flavone has shown significant antiviral effects on both HIV-1 reactivation and inhibition of Epstein-Barr virus (EBV) reactivation in cells. Besides these antiviral activities, luteolin also showed antiviral effects against Chikungunya virus, Japanese encephalitis virus [10], severe acute respiratory syndrome coronavirus (SARS-CoV) and rhesus rotavirus. Among flavonols the antiviral effect of quercetin has been most extensively investigated. Quercetin has been found to demonstrate an *in-vitro* dose-dependent antiviral activity against respiratory syncytial virus, poliovirus type 1, HSV-1 and HSV-2. It was also reported that quercetin has potential ability as a prophylactic against Ebola virus infection [11]. Kaempferol, another flavonol extracted from *Ficus benjamina* leaves have shown inhibitory activity against HCMV, HSV-1, HSV-2 and influenza A virus [12].

Fisetin a modified flavonol has shown to inhibit CHIKV infection as well as the HIV-1 infection by blocking viral entry and virus-cell fusion [12]. Resveratrol and pterostibenes have been found to exhibit anti-viral activities against a wide range of viruses including HIV-1 [13]. Resveratrol has been shown to inhibit the replication of pseudorabies virus (PVR) and Middle East Respiratory Syndrome coronavirus (MERS-CoV) and also reduced the expression of MERS-CoV nucleocapsid (N) protein [14-17]. Pterostilbene, a natural dimethylated analogue of resveratrol found in berries and grapes has been found to inhibit the replication of several viruses, including herpes simplex viruses (HSVs) 1 and 2, varicella-zoster virus, influenza virus and human papillomaviruses. Considering the contagiousness of the COVID-19 and its consequences, there is an imperative need to develop an efficient therapy to curtail the spread of this deadly virus and safely treating the infected individuals. In that direction, the repurposing of the FDA approved existing drugs like chloroquine (CQ) and HCQ either alone or in combination with other known drugs are currently being attempted [18, 19]. Preliminary *in vitro* studies and clinical trials carried out by scientists on COVID-19 patients disclosed the effectiveness of HCQ, an anti-malarial drug in combination with azithromycin, a broad spectrum anti-bacterial drug in reducing the viral load [20]. Although some of these initial studies disclosed promising results, a lot still remains to be done to analyse their compatibility, cost, accessibility, side effects, dosages etc. Currently scientists are indulging themselves to identify ideal natural compounds that can target and modulate unique or novel sites like the spike glycoprotein (S) on the surface of SARS-CoV-2 [21, 22]. In this *in-silico* study, we have performed molecular docking experiments to ascertain the most potent natural compounds (flavonoids) that can bind to the functional domains of the SARS-CoV-2S protein, a viral surface glycoprotein required for initial attachment and internalisation within host cells [1]. We found that about 10 of these compounds effectively binds to the C-terminal region of either the S1 domain or the S2 domain of SARS-CoV-2S and their binding interaction are more stable than with that of HCQ. These natural compounds are capable of binding to either the S1 or S2 domains of the SARS-CoV-2S protein and most probably prevent it from binding to the ACE receptor or internalization during fusion [23, 24]. The ADME analysis also suggested that most of these compounds have potential to function as effective anti-SARS-CoV-2 agents and needs further scientific explorations.

Material and methods:

Preparation of Ligands and Receptor:

The 3-dimensional structure of all ligands (Kaempferol, Curcumin, Pterostilbine, Hydroxychloroquine, Fisetin, Quercetin, Isorhamnetin, Genistein, Luteolin, Resveratrol and Apigenin) was downloaded from the PubChem database and then these structures were converted in pdb format by using PyMol [25]. The structure of SARS-CoV-2S protein (Fig. 1) was downloaded from the RCSB protein data bank (PDB-ID: 6VYB) [21]. Structure of all the ligands have been provided in table S1

Molecular docking of Phytochemicals on SARS-CoV-2 S (spike protein):

The cryo-electron microscopic structure of SARS-CoV-2S was used for the docking analysis. SARS-CoV-2S protein is a heterotrimer consisting of chain A, B and C [21]. For the docking experiment, chain A of the spike protein was used. First, the SARS-CoV-2S, Curcumin and HCQ were converted into pdbqt format using autodoc tools. Polar hydrogens and gasteiger charges were added to SARS-CoV-2S, curcumin and HCQ structure before docking. The molecular docking tool autodoc vina [26] was used to study the binding of curcumin and hydroxychloroquine on the SARS-CoV-2S. Further, blind docking of curcumin was performed to know the probable binding sites. For this, entire SARS-CoV-2S molecule was covered with the grid box of dimension 76 Å × 92 Å × 160 Å with grid spacing 1 Å. The SARS-CoV-2S was kept rigid while the curcumin was kept flexible. The four sets of docking were performed with exhaustiveness 100. Each set of autodoc vina produced 9 conformations, among them, 6 conformations were docked at one domain of SARS-CoV-2S, that domain is used for the local docking. For local docking, SARS-CoV-2S was covered with the grid box of dimension 60 Å × 54 Å × 66 Å with grid spacing 1 Å. The exhaustiveness was kept at 100. Four sets of local docking were performed for each ligand and the site where the maximum number of conformations bind was determined as a binding site. The conformations with high negative binding energy are represented in the figures. The docking results were analyzed using MGL tool 1.5.6 [27] and the hydrophilic and hydrophobic interactions were determined using PyMol [25].

Similar to curcumin, the docking of other ligand was performed on the SARS-CoV-2S protein. The docking parameters of all the ligand have been provided in table 1.

Prediction of ADME by computational analysis: ADME profiling of all the flavonoids at pH 7 were determined using online software tools [28]. The important parameters allied with ADME properties such as Lipinski's rule of five, solubility of drug, pharmacokinetics properties, molar refractivity and drug likeliness were deliberated [29]. All calculated values are provided in table 3.

Results and Discussion:

In the field of computer aided drug designing particularly for the identification of a lead compound [30, 31], molecular docking is immensely employed to explore the various types of binding interaction of the prospective drug with the different domains or active sites on the target molecules. Among all different types of interactions such as H-bond, π-π, amide-π interactions etc., the binding efficacy of a ligand molecule with the active sites of a target have widely been explained by evaluating its hydrogen bonding pattern [30, 32] and the nature of residues present at the active site. The binding energy (Kcal/mol) data allows us to study and compare the binding affinity of different ligands/compounds with their corresponding target receptor molecule. A lower binding energy indicates a higher affinity of the ligand for the receptor. The ligand with highest affinity can be chosen as the potential drug for further studies. The SARS-CoV-2S is a surface glycoprotein of 2019 novel corona virus. This protein plays important roles during viral attachment, fusion and entry into the host cells [33] . Biologically this protein exists in a heterotrimeric form with three separate polypeptide chains: chain A, B and C, forming each monomer [34]. For the present study, ten phytochemicals of flavonoids and non-flavonids class with broad range of biological activities along with one FDA-approved anti-malarial drug HCQ which have exhibited its efficacy against SARS-CoV-2 have been selected as ligands to investigate their binding affinity with SARS-CoV-2S chain A as the receptor target protein. The docking study revealed that out of the three different domains namely S1-N terminal, S1-C terminal and S2, these phytochemicals and HQC exhibit binding affinity for mainly two domains S1-C terminal and S2 of the spike protein of SARS-CoV-2. Among all the ligands, 2-(3,4-dihydroxyphenyl)-3,5,7-trihydroxy-4H-chromen-4-one (quercetin) (Fig. 2 and Table 2) and 2-(3,4-dihydroxyphenyl)-3,7-dihydroxy-4H-chromen-4-one (fisetin) (Fig. 3 and Table 2) has

displayed lowest and identical binding energy of -8.5 Kcal/mol as well as similar binding preferences for S2 domain of the spike protein. Despite of having similar preference for the S2 domain, the presence of additional 5-OH group on the chromone ring of querecetin affected its hydrogen bonding interactions compared to fisetin which also got reflected in their interaction with different residues of S2 domain. As shown in Fig. 3 and Table 2, the fisetin interacts with SER 730, THR 778 and HIS 1058 residues through H-bonding and exhibits hydrophobic interaction (based on 3D views different OH forming H-bonds and their bond length could have also been specified) with ILE 870, PRO 880 and THR 732 residues of S2 domain of the spike protein. Whereas, quercetin forms hydrogen bond with LYS 733, LEU 861, MET 731,SER 730,PRO 1057, GLY 1059, HIS 1058 and ALA 1056 residues and displays hydrophobic interaction with ILE 870, ASP 867, MET 730, VAL 860 and PRO 863. Similarly, the presence of one methoxy (OMe) group in case of 3,7-dihydroxy-2-(4-hydroxy-3-methoxyphenyl)-4*H*-chromen-4-one (isorhamnetin) (Fig. 4 and Table 2) which is a methyl ether of quercetin, displayed slightly lower binding affinity (-8.3 Kcal/mol). Compared to quercetin, replacement of –OH group at 3'-C of phenyl ring with sterically bulkier OMe group in isorhamnetin, led to the reduction in hydrogen bond donation ability and at the same time affected the nature of residues present at the interaction site (Table 2). Similarly, lack of one hydroxyl (-OH) group at 3'-C of phenyl ring in case of 3,5,7-trihydroxy-2-(4-hydroxyphenyl)-4*H*-chromen-4-one (kamferol) (Fig. 5 and Table 2) not only lead to the reduction in binding affinity (-7.4 Kcal/mol) but it also lead to binding at S1 domain rather than S2 domain which were observed in case of other three flavonols. Among two flavones (2-arylchromones), 2-(3,4-dihydroxyphenyl)-5,7-dihydroxy-4*H*-chromen-4-one (luteolin) with an additional OH group at 3'-C of phenyl ring exhibited better binding affinity (-8.2 Kcal/mol) compared to 5,7-dihydroxy-2-(4-hydroxyphenyl)-4*H*-chromen-4-one (apigenin) (Fig. S1 and Table 2) which displayed somewhat lower binding energy (-7.7 Kcal/mol) at S2 domain. 5,7-dihydroxy-3-(4-hydroxyphenyl)-4H-chromen-4-one (genistein) (Fig. S2 and Table 2) which is an isoflavone also exhibited good binding affinity (-8.2 Kcal/ mol) at S2 domain. We also found that luteolin binds with S2 domain (similar binding affinity like genistein (-8.2 Kcal/ mol) (Fig. S3 and Table 2). The docking studies performed with stilbenoids such as (*E*)-5-(4-hydroxystyryl) benzene-1, 3-diol (resveratrol) (Fig. S4 and Table 2) and (*E*)-4-(3,5-dimethoxystyryl)phenol (pterostilbene) (Fig. S5 and Table 2) revealed relatively better binding affinity of resveratrol (-7.9 Kcal/mol) compared to the later one i.e. pterostilbene

(-6.7 Kcal/mol). The reduction in binding affinity and change in binding site in case of pterostilbene (-6.7 Kcal/mol) compared to resveratrol (-7.9 Kcal/mol) could be attributed to the methylation of 2-hydroxyl groups in case of the earlier one which led to the decrease in hydrogen donor ability of resveratrol. The docking study performed with (1*E*,6*E*)-1,7-bis(4-hydroxy-3-methoxyphenyl)hepta-1,6-diene-3,5-dione (curcumin) (Fig. S6 and Table 2) which is an active ingredient present in turmeric, with the spike protein of SARS-CoV-2S also revealed average binding affinity (-7.1 Kcal/mol). It forms the hydrogen bonds with the GLN 564, ALA 520 and ASN 544 and exhibits hydrophobic interaction with THR 430, LEU 517, PHE 565, LEU 546, LEU 390 and VAL 382 at the S1 domain. Likewise the binding studies performed with hydroxychloroquine (HCQ) which has emerged as a potent therapeutic option against COVID-19 and received much attention in the last few days, revealed the lowest binding affinity (-5.6 Kcal/mol) among all others (Fig. 6 and Table 2) . It interacts with the ALA 520, GLN564, PHE 565, ARG 567 and HIS 519 residues through hydrogen bond formation. It forms the hydrophobic interaction with LEU 518, LEU 517, CYS 391, LEU 546 and ALA 522. The lowest binding affinity observed with HCQ could be attributed to the presence of confirmationally labile and sterically bulkier carbon chain which probably disrupts the interaction of the HCQ with the binding site leading to the reduced binding affinity. Overall the docking study discloses two different sets of ligands which bind at S1 and S2 domains of SARS-CoV-2S.

Lipinski rule of five is generally used to [28, 29] evaluate potential interactions between drug and other target non-drug molecules. It evaluates the propensity of a compound with a certain pharmacological or biological activity to be used as a potential drug. It acts like a filter to screen potential therapeutic agents/drugs just at the initiation of the programme, thereby minimizing the labor and cost of exercises involving clinical drug development and to a large extent preventing late stage clinical failures. The rule mainly determines the different molecular properties of a compound which are its prime requisition to be a potential drug like absorption, distribution, metabolism and excretion (ADME). Lipinski's rule states that for any compound to be selected as a potential drug it should have (a) Molecular mass < 500 Dalton (b) high lipophilicity (expressed as LogP$\leq$5) (c) less than 5 hydrogen bond donors (d) Less than 10 hydrogen bond acceptors (e) molar refractivity between 40-130. If a compound of interest possesses more than two of the aforementioned criteria then the compound is likely to be a potential candidate for drug development. All the phytochemicals used in this study were found to pass all the five

criteria's mentioned above in Lipnaski's rule (Table 3). Thus we suggest that all of these phytochemicals have the potential ability to work effectively as novel drugs.

The results of our entire article are based on virtual computational screening. We wanted to share our findings with all the scientists working in the field of anti-SARS-CoV-2 research throughout the globe as early as possible for experimental drug verification. *In vitro* and *in vivo* experiments with our selected group of potential anti-viral compounds will strengthen our perspectives that natural products based therapeutic interventions is the need of the hour. Herbal medicines formulated from the phytochemicals we predicted, will be extremely essential for the treatment of SARS-CoV-2 infections.

Conclusion

This *in-silico* approach to find a novel natural compound that binds and prevents the attachment/internalization of SARS-CoV-2 is decisively a perfect therapeutic option in this period of time constraint. Bioinformatics has been effectively used by many studies to make a fast and more or less accurate prediction of potential drugs or inhibitors. In this study, we have used multiple bioinformatics tools to identify potent natural compounds mainly flavonoids that targets and binds to the spike protein of SARS-CoV-2. We have identified about 10 flavonoids out of the whole pool capable of binding either to the S1 or S2 domain of the SARS-CoV-2S protein. Our findings suggest that all of these compounds have the potential to inhibit SARS-CoV-2 and should be explored further as preventive therapeutics for COVID-19.

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Table 1:

S. No.	Ligand	Docking Parameters	
		Blind Docking	**Local Docking**
1	Kamferol	96Å × 96 Å × 160 Å	58 Å × 56 Å × 72 Å
2	Curcumin	76 Å × 92 Å × 160 Å	60 Å × 54 Å × 66 Å
3	Pterostilbene	96Å × 96 Å × 160 Å	58 Å × 56 Å × 72 Å
4	Hydroxychloroquine	76 Å × 92 Å × 160 Å	58 Å × 56 Å × 72 Å
5	Fisetin	84 Å × 126 Å × 160 Å	40 Å × 44 Å × 118 Å
6	Quercetin	96Å × 96 Å × 160 Å	40 Å × 44 Å × 118 Å
7	Isorhamnetin	96Å × 96 Å × 160 Å	40 Å × 44 Å × 118 Å
8	Genistein	146 Å × 126 Å × 160 Å	40 Å × 44 Å × 118 Å
9	Luteolin	96Å × 96 Å × 160 Å	40 Å × 44 Å × 118 Å
10	Resveratrol	96Å × 96 Å × 160 Å	40 Å × 44 Å × 118 Å
11	Apigenin	126 Å × 102 Å × 160 Å	44 Å × 96 Å × 98 Å

Table 1: The Docking parameters used for different ligands.

Table 2:

Compound	Binding Affinity (kcal/mol)	Interacting Domain of Spike Protein	Interacting amino acid Residue
Kamferol	-7.4	C-Terminal of S1 Domain	THR 393, ALA 522, LEU 527, CYS 391, LEU 390, ASN 544, PHE 782, ALA 1056
Curcumin	-7.1	C-Terminal of S1 Domain	THR 430, LEU 517, ALA 520, GLN 564, PHE 565, ASN 544, LEU 546, LEU 390, VAL 382
Pterostilbene	-6.7	C-Terminal of S1 Domain	THR 393, ALA 522, ALA 520, HIS 519, ASN 544, GLN 564, LEU 390, GLY 545, PHE 543, LEU 546,PHE 565
Hydroxychloroquine	-5.6	C-Terminal of S1 Domain	GLN564, PHE 565, ALA 520, ARG 567, HIS 519, LEU 518, LEU 517, CYS 391, LEU 546, ALA 522
Fisetin	-8.5	S2 Domain	ILE 870, PRO 880, SER 730, HIS 1058, THR 732, THR 778
Quercetin	-8.5	S2 Domain	ILE 870, ASP 867, ALA 1056, PRO 1057, GLY 1059, HIS 1058, SER 730, MET 730 MET 731, LYS 733, VAL 860, LEU 861, PRO 863

Isorhamnetin	-8.3	S2 Domain	HIS 1058, VAL 729, SER 730, MET 731, THR 732, LYS 733, VAL 860, LEU 861, PRO 863, THR 778, ILE, 870, PHE 782, ALA 1056
Genistein	-8.2	S2 Domain	GLY 1059. SER 730, HIS 1058, THR 732, LYS 733, PRO 863, ASN 867, THR 870, PHE 782, ALA 1056
Luteolin	-8.2	S2 Domain	ALA 1056, GLY 1059, HIS 1058, ASP 867, ILE 870, THR 871, PRO 863, LEU 861, LYS 733, MET 731
Resveratrol	-7.9	S2 Domain	THR 732, HIS 1058, LYS 733, LEU 861, PRO 863, ASP 867, ILE 870, PHE 782
Apigenin	-7.7	S2 Domain	SER 730, HIS 1058, ALA 1056, ILE 870, PRO 863, LEU 861

Table 2: Molecular docking analysis to find out the putative binding sites of selected inhibitors on SARS-CoV-2S (spike protein)

Table 3:

S.No.	Ligand/Phytochemicals	ADME Properties (Lipinki's Rule of Five		Drug Likeliness
		Properties	**Values**	
1.	Kamferol	Molecular weight (<500 Da)	286	Yes
		LogP (<5)	2.3	
		H-bond donar (5)	4	
		H-bond acceptor (<10)	6	
		Molar Refractivity (40-130)	72.4	
		Violations	**NO**	
2.	Curcumin	Molecular weight (<500 Da)	368	Yes
		LogP (<5)	3.4	
		H-bond donar (5)	2	
		H-bond acceptor (<10)	6	
		Molar Refractivity (40-130)	102	
		Violations	**NO**	
3.	Pterostilbene	Molecular weight (<500 Da)	256	Yes
		LogP (<5)	3.6	
		H-bond donar (5)	1	
		H-bond acceptor (<10)	3	
		Molar Refractivity (40-130)	76.6	
		Violations	**NO**	
4.	Hydroxychloroquine	Molecular weight (<500 Da)	335.5	Yes
		LogP (<5)	3.8	
		H-bond donar (5)	2	
		H-bond acceptor (<10)	4	
		Molar Refractivity (40-130)	98.3	
		Violations	**NO**	
5.	Fisetin	Molecular weight (<500 Da)	286	Yes
		LogP (<5)	2.3	
		H-bond donar (5)	4	
		H-bond acceptor (<10)	6	
		Molar Refractivity (40-130)	72.4	
		Violations	NO	

S.No.	Ligand/Phytochemicals	ADME Properties (Lipinki's Rule of Five		Drug Likeliness
		Properties	**Values**	
6.	Quercetin	Molecular weight (<500 Da)	302	Yes
		LogP (<5)	2	
		H-bond donar (5)	5	
		H-bond acceptor (<10)	7	
		Molar Refractivity (40-130)	74	
		Violations	**NO**	
7.	Isorhamnetin	Molecular weight (<500 Da)	316	Yes
		LogP (<5)	2.3	
		H-bond donar (5)	4	
		H-bond acceptor (<10)	7	
		Molar Refractivity (40-130)	79	
		Violations	**NO**	
8.	Genistein	Molecular weight (<500 Da)	270	Yes
		LogP (<5)	2.4	
		H-bond donar (5)	3	
		H-bond acceptor (<10)	5	
		Molar Refractivity (40-130)	71	
		Violations	**NO**	
9.	Luteolin	Molecular weight (<500 Da)	286	Yes
		LogP (<5)	2.1	
		H-bond donar (5)	4	
		H-bond acceptor (<10)	6	
		Molar Refractivity (40-130)	72.5	
		Violations	**NO**	
10.	Resveratrol	Molecular weight (<500 Da)	228	Yes
		LogP (<5)	3	
		H-bond donar (5)	3	
		H-bond acceptor (<10)	3	
		Molar Refractivity (40-130)	67	
		Violations	NO	
11.	Apigenin	Molecular weight (<500 Da)	270	Yes
		LogP (<5)	2.4	
		H-bond donar (5)	3	
		H-bond acceptor (<10)	5	
		Molar Refractivity (40-130)	71	
		Violations	NO	

Table 3: ADME Properties of selected inhibitors against SARS-CoV-2S (spike protein)

Figure legends

Figure1. The structure of SARS-CoV-2S (spike protein) (PBD-ID: 6YVB) (A) FASTA sequence of the SARS-CoV-2S (spike protein) chain A (B & C) The cartoon showing the structure and surface of SARS-CoV-2S (spike protein) chain A.

Figure 2. The molecular docking of SARS-CoV-2S and Quercetin. (A) The 3- dimensional ribbon structure of SARS-CoV-2S protein. (B) The 3-dimensional structure of quercetin. (C) The putative binding site of quercetin on SARS-CoV-2S protein. (D) The interacting amino acid residues of SARS-CoV-2S with quercetin.

Figure 3. The molecular docking of SARS-CoV-2S and fisetin. (A) The 3- dimensional ribbon structure of SARS-CoV-2S protein. (B) The 3-dimensional structure of fisetin. (C) The putative binding site of fisetin on SARS-CoV-2S protein. (D) The interacting amino acid residues of SARS-CoV-2S with fisetin.

Figure 4. The molecular docking of SARS-CoV-2S and isorhamnetin. (A) The 3 dimensional ribbon structure of SARS-CoV-2 S protein. (B) The 3-dimensional structure of isorhamnetin. (C) The putative binding site of isorhamnetin on SARS-CoV-2S protein. (D) The interacting amino acid residues of SARS-CoV-2S with isorhamnetin.

Figure 5. The molecular docking of SARS-CoV-2S and kaempferol. (A) The 3- dimensional ribbon structure of SARS-CoV-2S protein. (B) The 3-dimensional structure of kaempferol. (C) The putative binding site of kaempferol on SARS-CoV-2S protein. (D) The interacting amino acid residues of SARS-CoV-2S with kaempferol.

Figure 6. The molecular docking of SARS-CoV-2S and hydroxychloroquine. (A) The 3-dimensional ribbon structure of SARS-CoV-2S protein. (B) The 3-dimensional structure of hydroxylchloroquine. (C) The putative binding site of hydroxychloroquine on SARS-CoV-2S protein. (D) The interacting amino acid residues of SARS-CoV-2S with hydroxychloroquine.