IN SILICO METHOD FOR SEARCHING POTENTIAL INHIBITORS OF SOUTHEAST ASIAN REGIONAL MEDICINAL PLANTS AGAINST THE MAIN PROTEASE STRUCTURE OF SARS COV-2 VIA AUTODOCK VINA

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Abstract- **Southeast Asian medicinal herbs with physiological action that include phytochemicals were examined for drug-like characteristics.20 phytochemicals were selected from 178 phytochemicals that were evaluated using the LIPINSKI rule of five and swiss-ADME. The spike proteins of the cov-2 variants were docked with these phytochemicals using Autodock Vina. Linalool taken as a standard in docking analysis, exhibited a binding affinity of -4.5 kcal/mol , -4.1 kcal/mol and -4.6 kcal/mol with respect to three cov-2 SARS spike variant respectively. Among the 60 docked result most alocasia and apigenin flavone glucoside shows very good docking scores. Alocasin B and vitexin produced a highest docking score of -10.1 kcal/mol and -10.3 kcal/mol with respect the spike protein of 6LU7. Alocasin B and Campesterol shows highest docking score of -8.3 kcal/mol and -8.3 kcal/mol with the spike protein of 6LZG. Campesterol and Alpha-muurolol produced a highest binding affinity score of -10.7 kcal/mol and -7.8 kcal/mol with the spike protein of 6M0J and are the prospective candidates for antiviral medications that might be used to treat the SARS-CoV-2 spike variants.**

Keywords: **SARS CoV-2, LIPINSKI Rule, swiss-ADME, phytochemicals, molecular docking.**

1. INTRODUCTION:

An annual herb, Perilla frutescens (L.) Britt., is a member of the Lamiaceae family of mints [1,2]. It is also known as beefsteak plant, purple mint, perilla mint, Chinese basil, Korean perilla, zisu in China, shiso in Japan, and tia to in Vietnam [3]. Perilla is the most frequent name for it. In the Japanese Pharmacopoeia (1991), Herba Perillae is described as a medication made from the leaves and twigs of perilla, but "Perilla leaf," "Perilla stalk," and "Perilla seed" correlate to Folium Perillae, Caulis Perillae, and Fructus Perillae in the Chinese Pharmacopoeia (1990) [4]. In Asian societies, it has long been utilized as both a traditional remedy and a useful food [5]. Due to the fact that the flower emerges from the inflorescence spadix, plants belonging to the araceae family are categorized as monocotyledonous flowering plants. Over 3,700 species belonging to 107 genera make up the Araceae family, which has a global distribution (Erlinawati, 2010). The Alocasia genus, which presently has 113 species and 2 species that have not yet been described, is the biggest in the family (Nauheimer et al., 2012). 79 species are listed as native to tropical and subtropical Asia, which stretches from the subtropical eastern Himalayas to India, China, Japan, the Malay Archipelago, and till Oceania (Nauheimer et al., 2012; GBIF, 2019), of the 96 approved names of Alocasia species in total[6]. One of the most significant edible legume crops is the mung bean (Vigna radiata L.), which is cultivated on more than 6 million hectares globally (about 8.5% of the world's pulse area) and is mostly consumed in Asian homes. The mung bean is widely grown in many Asian countries (concentrated mainly in China, India, Bangladesh, Pakistan, and some Southeast Asian countries), as well as in dry regions of southern Europe and warmer parts of Canada and the United States, due to its characteristics of being a relatively drought-tolerant, low-input crop, and short growth cycle (70 days or so) [7].

BIOVIA Discovery Studio assessed the interactions between the ligands and proteins [8]. A computer procedure called molecular docking looks for the interaction between a large molecule (the receptor) and a tiny one (the ligand). Molecular docking is extremely valuable to drug design because it can be used to anticipate the binding conformations and affinities between drug molecules and their target proteins, helping to understand the biological process behind those binds [9]. The situation will get more complex if

flexibility is addedto the ligand or, further, to the receptor [9,10]. The complexity of modeling the molecular system is mostly caused by the high degree of flexibility.

The same organization that created the earlier iterations of AutoDock, one of the most well-known docking software, launched AutoDock Vina [11] (referred to as Vina thereafter). Vina use an empirical scoring system and an iterated local search global optimizer to assess the binding affinity between compounds. According toreports, this combination is able to outperform AutoDock 4 in terms of speed by almost two orders of magnitude while also providing substantially more accuratebinding mode predictions [11].

2. MATERIAL AND METHODS:

2.1: PREPARATION OF PROTEINS:

The modelled protein structures were prepared for docking using BIOVIA discovery studio. COVID-19 main protease in complex with an inhibitor N3 (PDB ID: 6LU7), SARS-CoV-2 spike receptor-binding domain bound with ACE2 (PDB ID: 6M0J), and spike receptor-binding domain complexed with its receptor ACE2 (PDB ID: 6LZG) and four spike proteins of SARS-CoV-2 spike variants were used to examine the interactions of the major phytocompounds of Mung bean, Thai basil, alocasia species, and perilla frutescens. Selected target proteins' three-dimensional structures were downloaded from the Protein Data Bank (PDB) at http://www.rscb.org/pdb. The target receptor molecule was given a hydrogen atom and non-essential water molecules, including heteroatoms, were taken out of it. Both of COVID-19's target proteins, SARS-CoV-2 spike protease (PDB ID: 6LU7) and SARS-CoV-2 spike receptor-binding domain (PDB ID: 6LZG), have binding sites for them. Grid box generation was used to identify the receptor binding (PDB ID: 6M0J). Structures were stored in pdbqt format and a grid box was created by alteringthe grid parameter's x, y and z coordinate values [12].

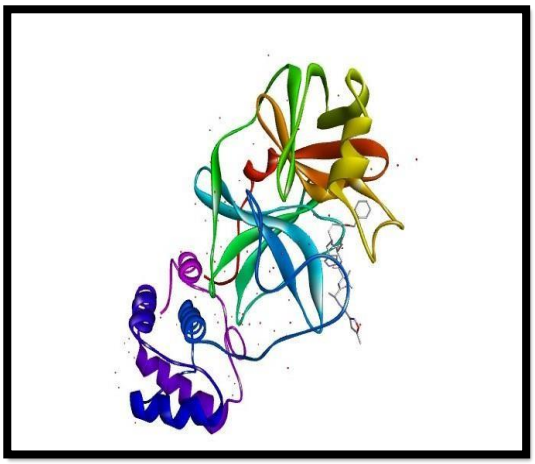


Fig:1 structure of 6LU7

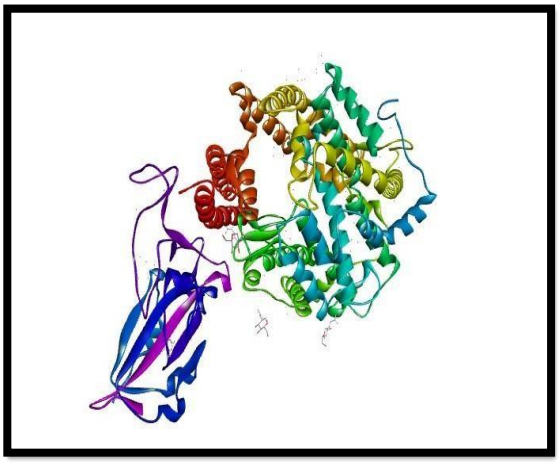


Fig:2 structure of 6MOJ

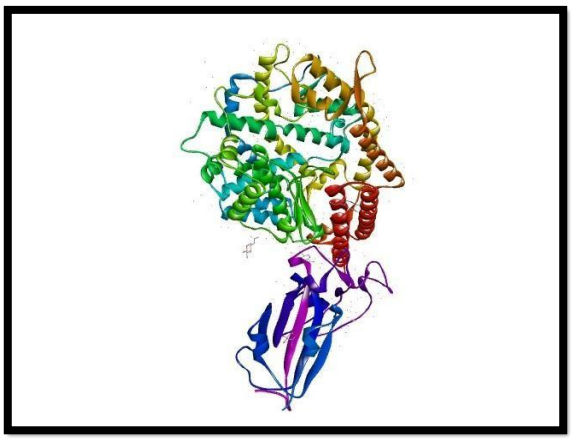


Fig:3 structure of 6LZG

2.2: PREPARATION OF LIGANDS:

Twenty bioactive compounds from different plant sources collated from public database and published research papers were downloaded from https://pubchem.ncbi.nlm.nih.gov in SDF format (Fig. 4). . These ligands

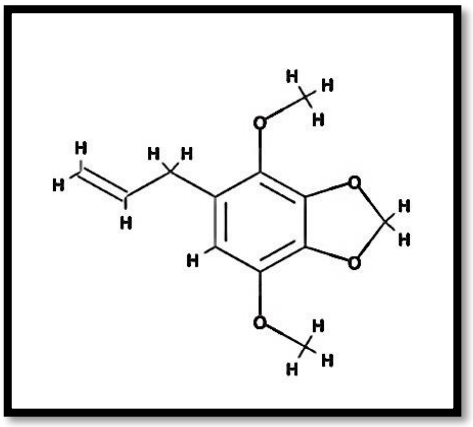
were converted to PDB format using the open babel program (http:// openbabel.org/wiki/Main Page), making them suitable for docking study. The torsion was then set after that settings for good binding with Autodock vina [15].

2.3: LIPINSKI'S RULE:

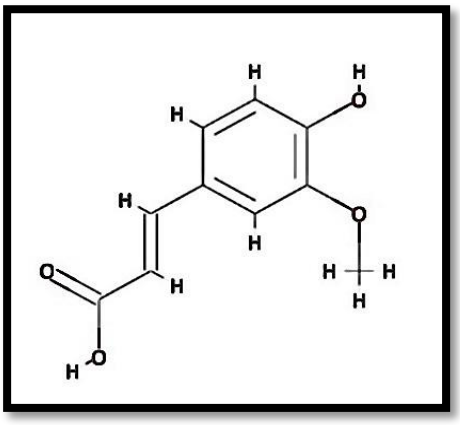
To assess a chemical's drug likeness a crucial stage in the drug development process Lipinski's Rule of five is utilized. This test helps researchers identify whether a given compound is likely to be orally active. Using the supercomputer capability for bioinformatics and computational biology, ligands were tested for the RO5 in this work. (http://www.scfbio-iitd.res.in/software/drugdesign/lipinski.jsp) [13, 14]. This investigation used bioactive substances with binding energies equivalent to those of common medications.

2.4: IN SILICO ADME ANALYSIS:

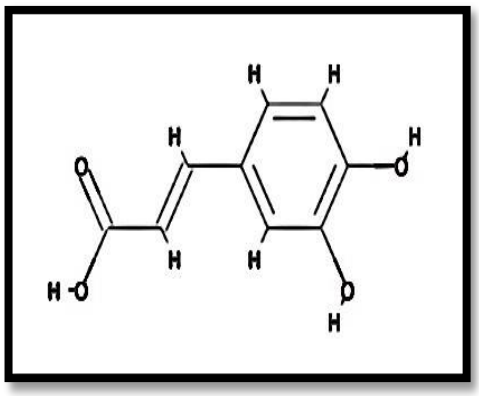
Absorption, Distribution, Metabolism, and Excretion are pharmacokinetics parameters that Swiss-ADME evaluation in the ligands (http://www.swissadme.ch/index.php). This assay's main goal is to offer information that will aid in the development of new drugs.



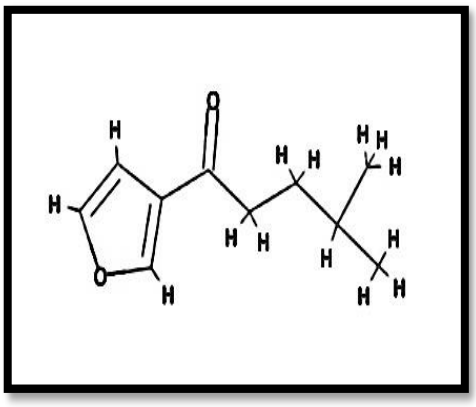
1. Apiol



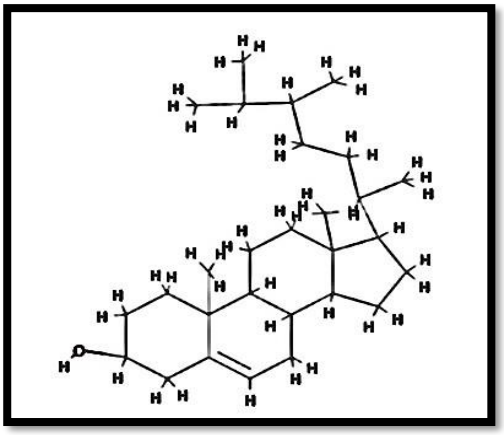
2. Ferulic acid

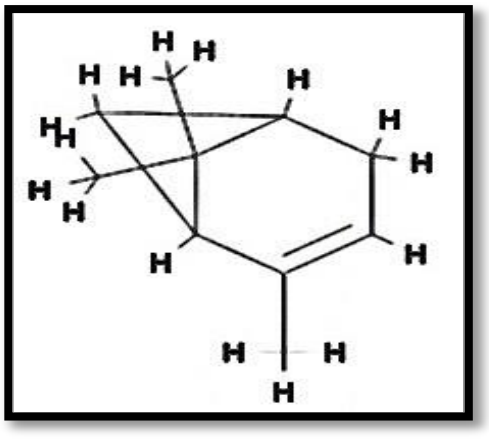


3.Caffic acid

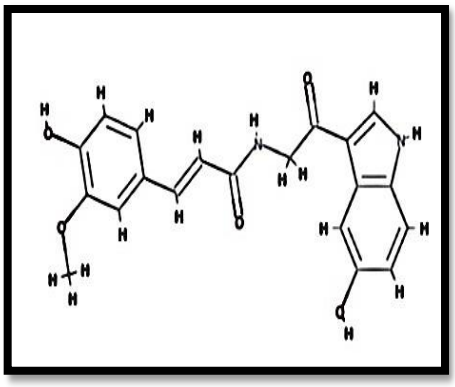


4. Perilla ketone

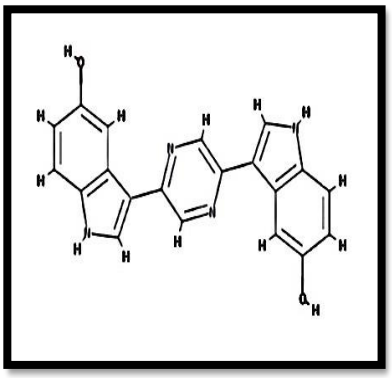




5. Campesterol

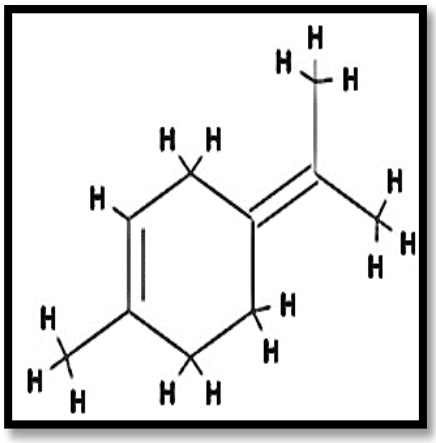


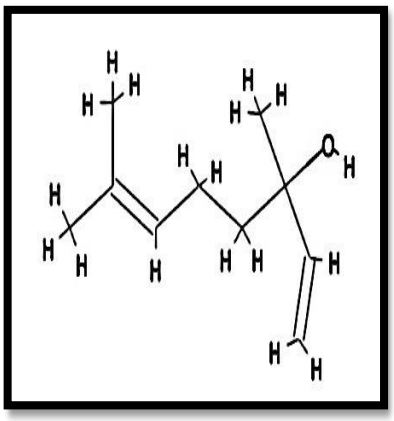
6. Alocasin A



7. Alocasin B

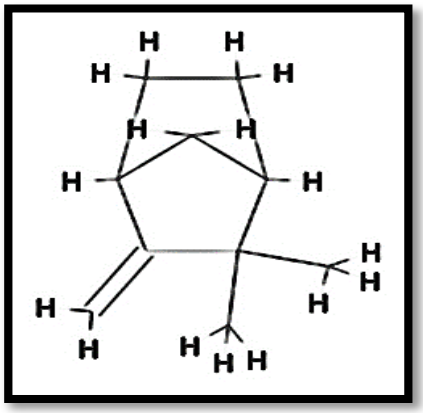
8. Alpha-pinene

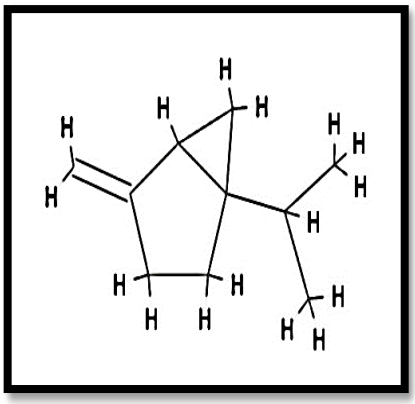




9. Terpinolone

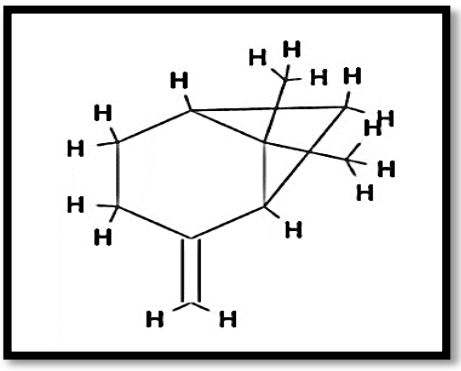
10. Linolool

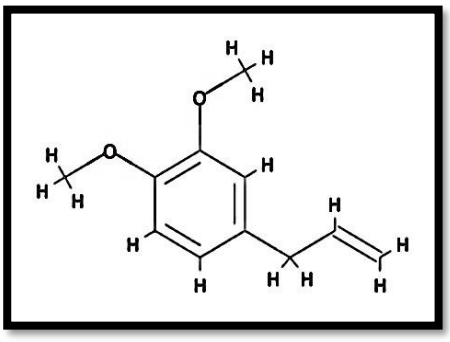




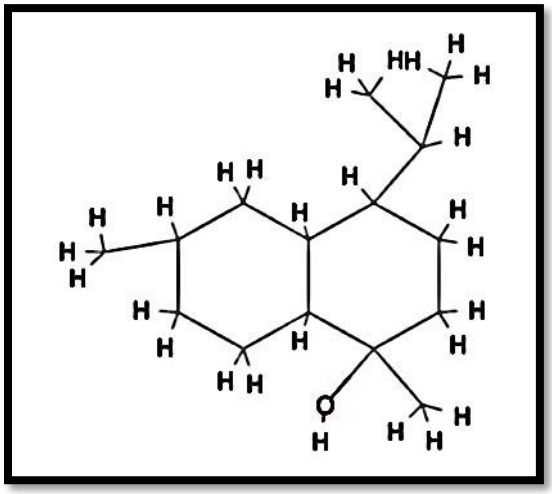
11. Camphene

12. Sabinene

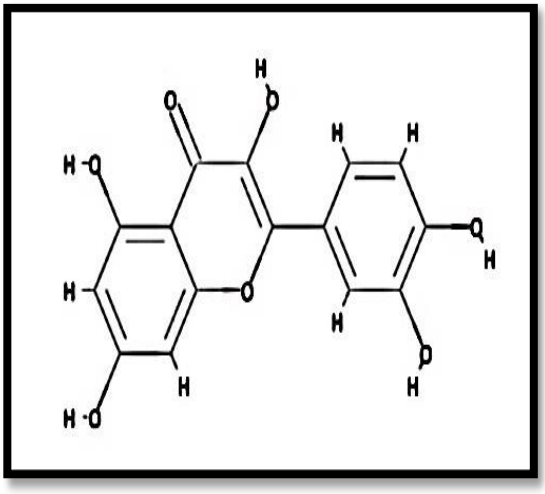




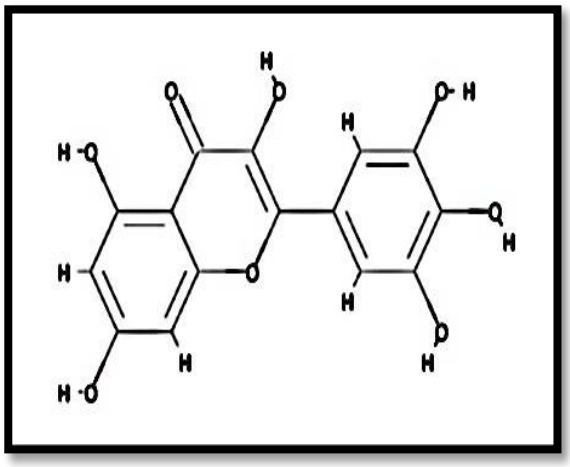
13. Beta-pinene



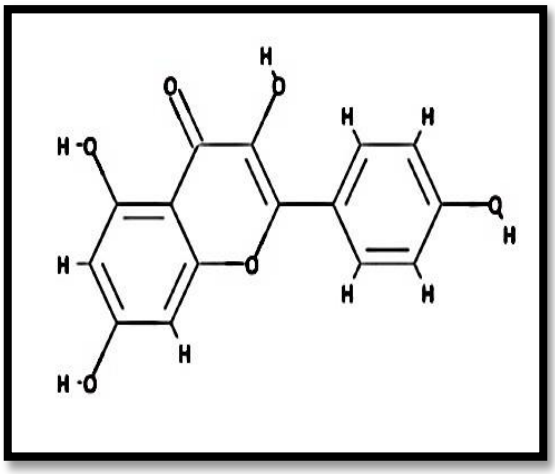
14. Methyl eugenol



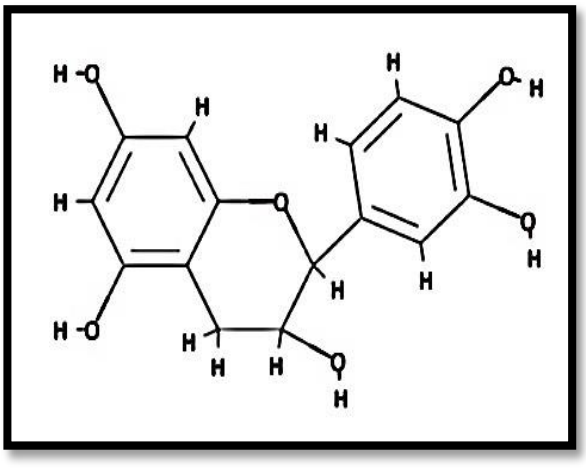
15. Alpha-muurolol



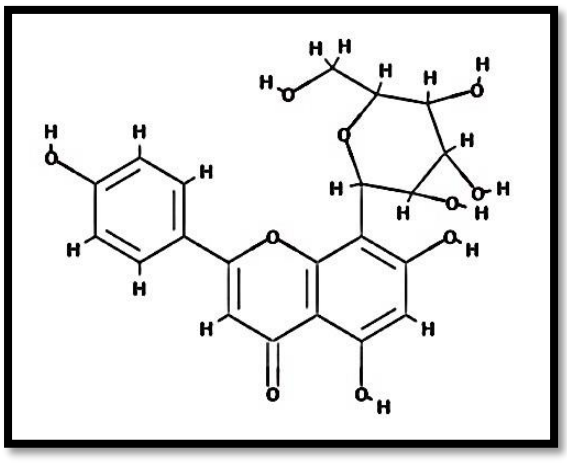
16. Quercetin



17. Myricetin



18. Kaempferol



19. Catechin

20. Vitexin

Fig. 4: Structures of 20 phytochemical compound

3.5: AUTODOCK VINA:

S.No	Ligands	Binding energy with SARS-spike protein (6LU7)	Binding energy with SARS-spike protein (6LZG)	Binding energy with SARS-spike protein (6M0J)
1.	Alocasin A	-7.5	-7.5	-6.5
2.	Alocasin B	-10.1	-8.3	-3.7
3.	Campesterol	-7.5	-8.3	-10.7
4.	Catechin	-8.4	-7.5	-6.6
5.	Kaempferol	-8.1	-7.7	-6.6
6.	Quercetin	-8.6	-7.8	-6.7
7.	Myricetin	-8.7	-7.9	-6.7
8.	Vitexin	-10.3	-8.2	-7.2
9.	Apiol	-6.0	-5.6	-6.0
10.	Caffic acid	-6.6	-6.0	-5.8
11.	Ferulic acid	-6.3	-6.8	-6.3
12.	Perilla ketone	-5.1	-5.6	-5.5
13.	Alpha-pinene	-4.8	-4.7	-4.5
14.	Camphene	-5.1	-4.9	-4.7
15.	Sabinene	-5.2	-5.0	-5.2
16.	Beta-pinene	-5.3	-5.7	-4.7
17.	Terpinolene	-5.3	-5.1	-4.3
18.	Alpha-muurolol	-7.2	-7.2	-7.8
19.	Methyl eugenol	-6.1	-5.7	-5.6
20.	Linolool	-4.5	-4.1	-4.6

Table 1: 20 phytochemicals docked with targeted spike protein of SARSCOV-2

Autodock Vina was used to conduct docking investigations. In the active site of the spike glycoprotein, a grid box of 62 68 40 was assigned. The 20 ligands in the collection were screened. The drug alocasin B served as a benchmark for contrasting docking scores. The docking outcomes are shown in **Table 1** was examined using the Discovery Studio Visualizer.Biovia, Dassault Systems, 2021). Moreover, the docking tool attach the 20 ligands into the spike proteins'6M0J receptor binding domain. Top 3 ligands were enlisted in **Table 2**[16].

S.no	Top 3 ligand	Binding affinity in 6LU7	Binding affinity in 6LZG	Binding affinity in 6M0J
1	Alocasin B	-10.3	-8.3	-3.7
2	Campesterol	-7.5	-8.3	-10.7
3	Vitexin	-10.3	-8.2	-7.2

Table 2: Top three ligand

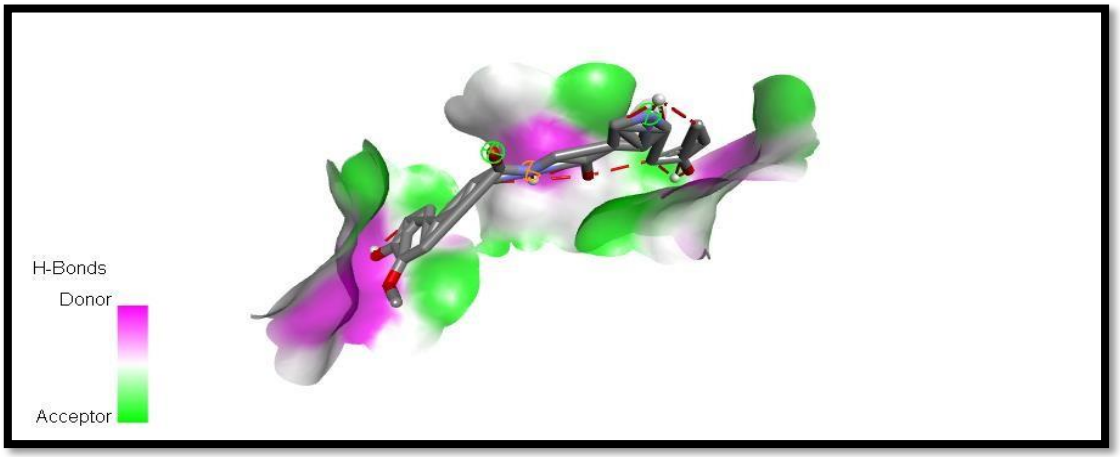
3. RESULTS AND DISCUSSION

178 phytochemicals that were initially chosen for this study came from 12 medicinal plants and were said to have anti-oxidant, anti-viral (or) anti- microbial effects. These phytochemicals were screened for drug resemblance using the LIPINSKI rule of five and SWISS-ADME. After much deliberation, 20 phytochemicals were chosen to

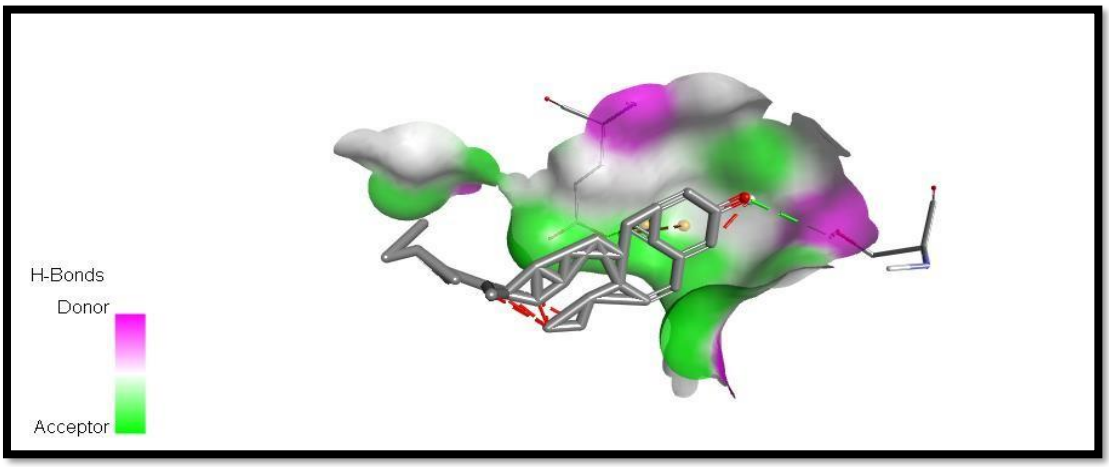
dock with the target spike protein of SAR-CoV-2.

Campesterol with a binding affinity of -7.5 kcal/mol , -8.3 kcal/mol and -10.7 kcal/mol with the spike protein variants (6LU7,6LZG and 6M0J). **Table 2** lists the top 3 phytochemicals by binding score, and they are all flavones. With justa few exceptions, the phytochemicals' binding affinities to the two targets arefairly comparable.

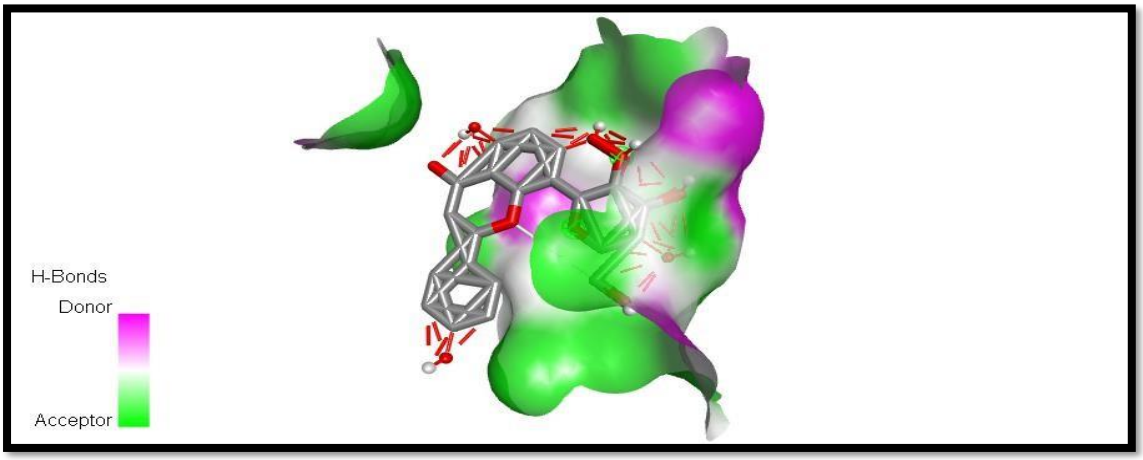
Among the top 3 hits, alocasin B, campesterol and vitexin have a more similarities and have more binding energy. . A344, A346, A347, A348 and B165are the most frequent amino acid residues at the binding site in all of these ligands.



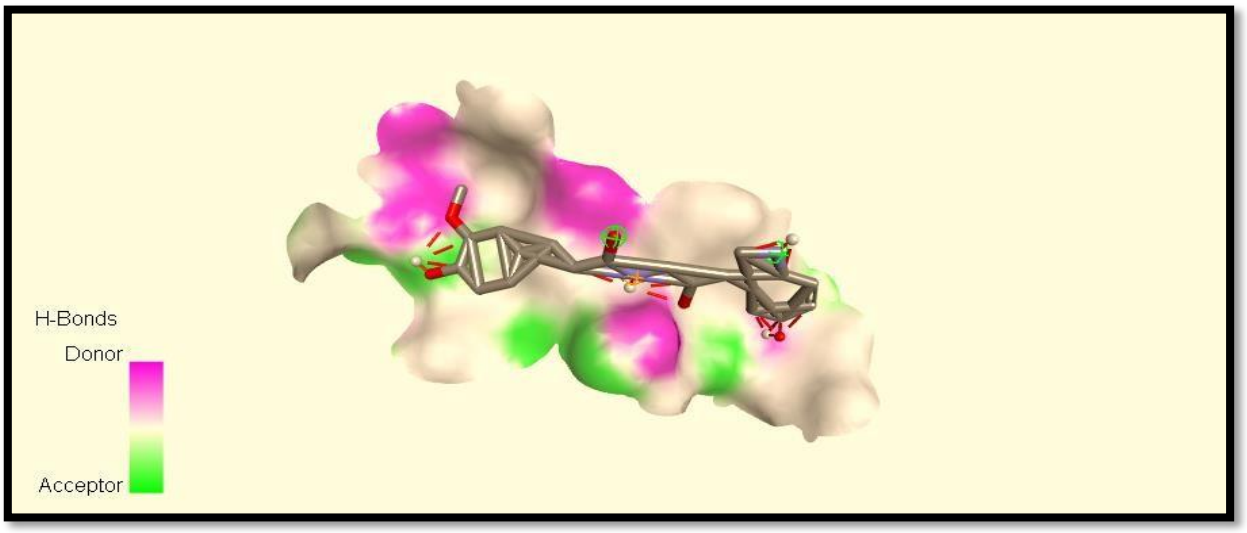
Alocasin B



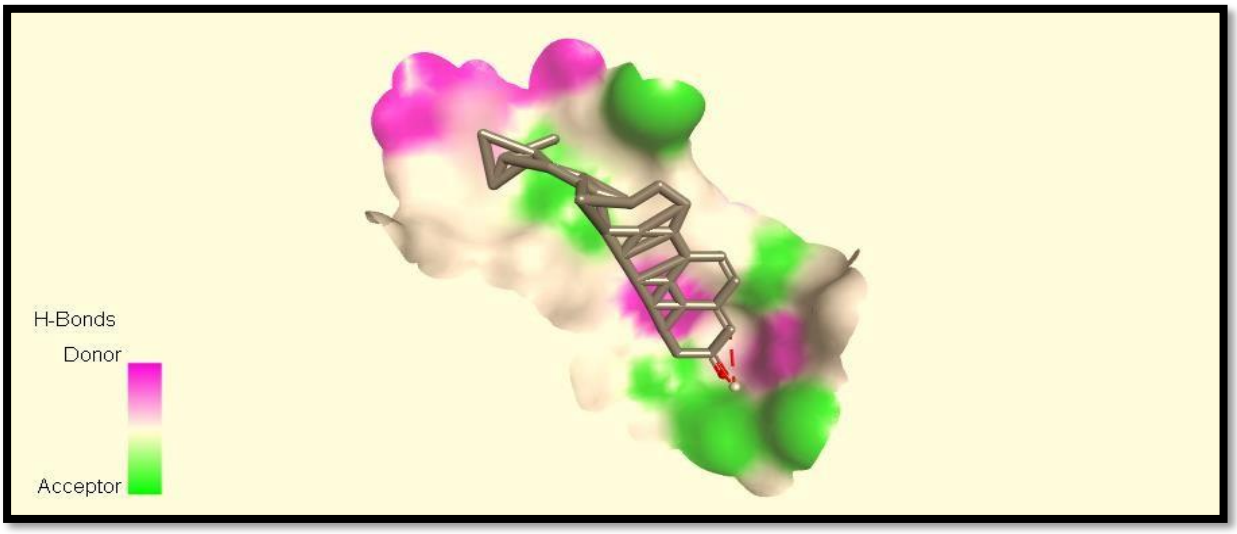
Campesterol



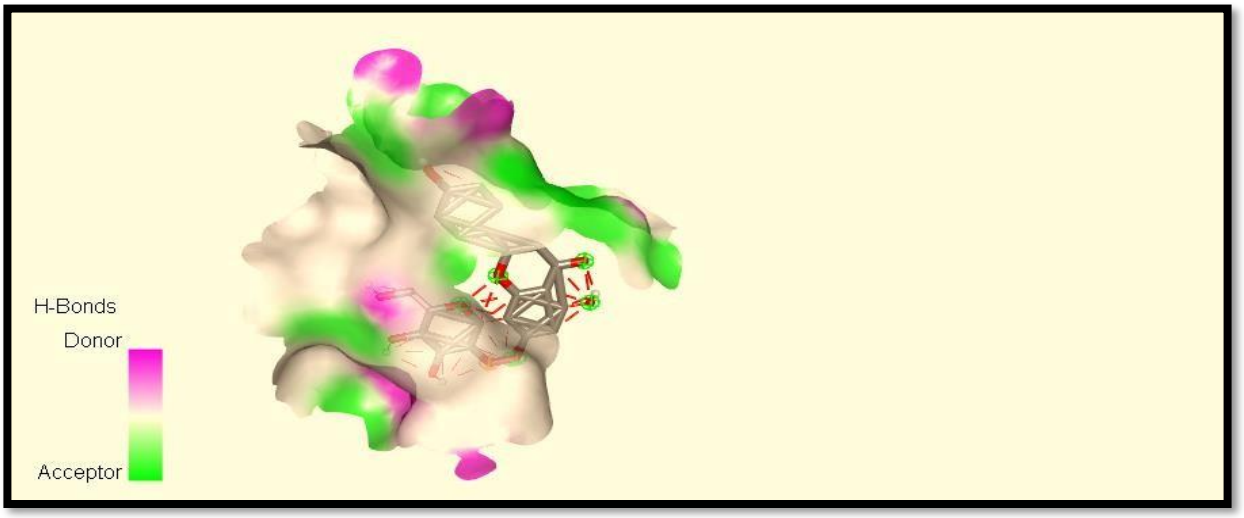
Vitexin



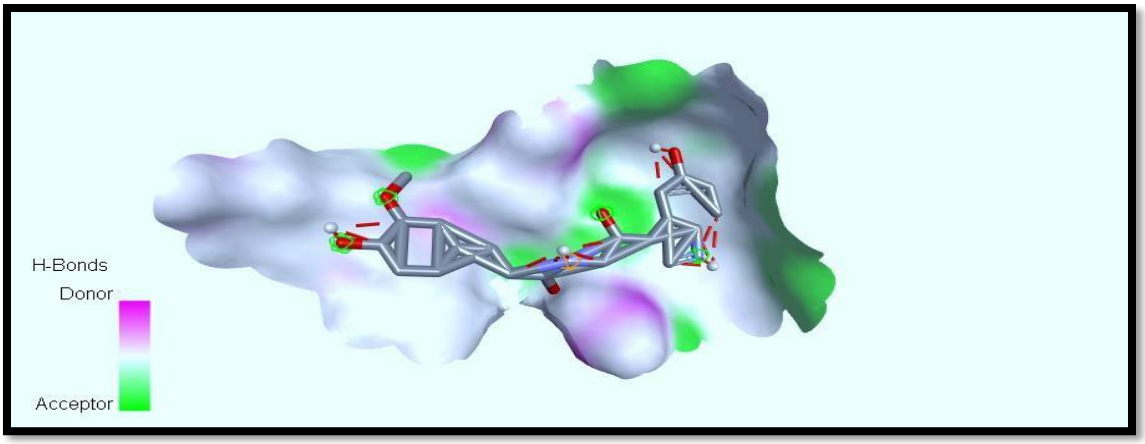
Alocasin B



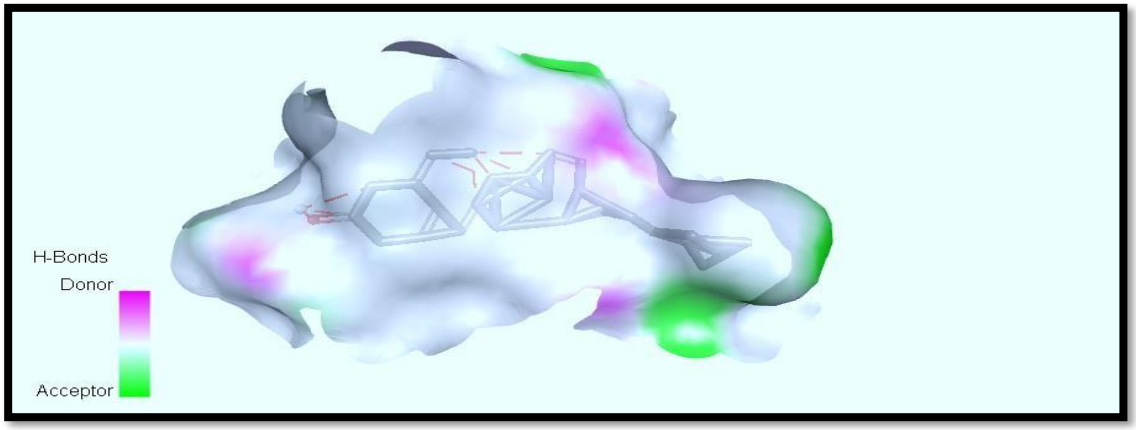
Campesterol



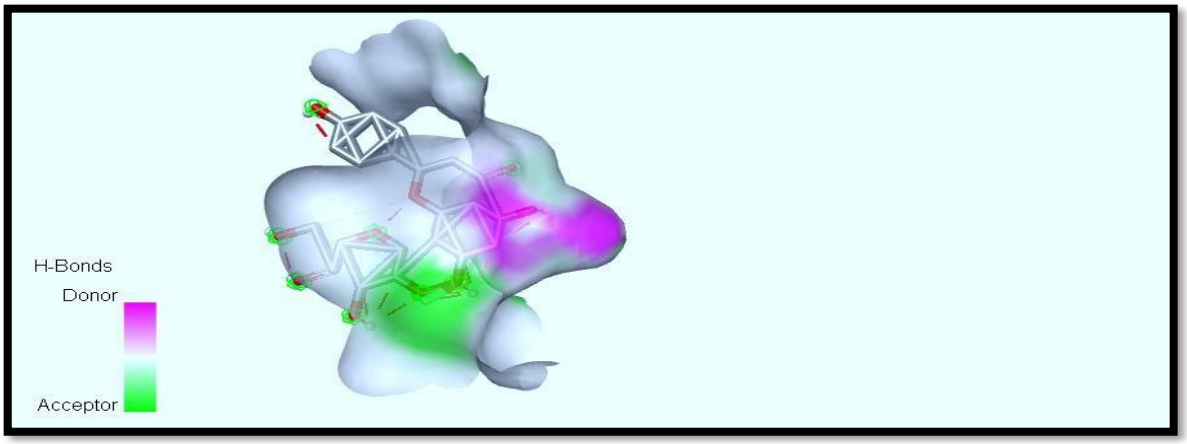
Vitexin



Alocasin B



Campesterol



Vitexin

Fig .5: Shows top 3 ligand interaction on 6LU7, 6LZG and 6M0J spike proteinsrespectively

The residues of the amino acids A344, A347, and A348 are engaged in interactions with the two aromatic rings of the ligands that are Pi-Pi T-shaped and Pi-alkyl. The involvement of the amino acid sequence A346 in B165 creates a hydrogen link with the aromatic hydroxyl instead of the usual hydrogen bond with the carbonyl oxygen between the aromatic rings group. The RBD has every common amino acid residue that is actively involved in interacting with the top hit ligands, which suggests that the ligands may be able to interfere with viral binding to the ACE2 receptor. The three aromatic pockets and the five acceptor groups all work together to connect with the spike proteins' active sites[17].

4. CONCLUSION:

The potential of several phytochemicals is investigated in an in silico study to analyze the antiviral action on spike variations. Twenty phytochemicals have drug-like and pharmacokinetics properties, according to our research. The pharmacodynamics and kinetic characteristics of these phytochemicals need to be determined, as well as the mechanism of action of these phytochemicals as nanoparticle carriers of antiviral drugs for successful therapy.

Acknowledgements

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