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In silico exploration of various Phytochemicals present in *Ficus hispida* Stem Bark as Hepatoprotective agents

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ABSTRACT

Ficus hispida Linn. is a moderate sized tree found throughout the year and is grown wild or cultivated for its edible fruits and folklore value. Traditionally, different parts of the plant have been used in the treatment of hemorrhage, diabetes, convulsion, hepatitis, dysentery, biliousness, lactagogue, and purgative. It contains wide varieties of bioactives from different phytochemical groups like alkaloids, carbohydrates, proteins and amino acids, sterols, phenols, flavonoids, gums and mucilage, glycosides, saponins, and terpenes. The main objective of this study was to explore the potential of various phytoconstituents [2-demethoxytylophorine **(1)**, lupeol acetate **(2)**, *n*-triacontanol **(3)**, oleanolic acid **(4)**, hispidin **(5)**, terpeneol **(6)**, β-amyrine **(7)**, linalool **(8)**, gluanol **(9)**, β-sitosterol **(10)**, and bergaptine **(11)**] as hepatoprotective agent by using *in-silico* approaches with the help of published literature on downregulation of enzyme/molecular component expression and combining this information in order to recognize drug target [PDB ID: 1IL6; Human Interleukin-6, NMR, Minimized Average Structure; https://doi.org/10.2210/pdb1IL6/pdb]. The *in silico* studies revealed that the phytoconstituents successfully inhibited the biological target at varied degree which suggested plausible utilization as hepatoprotectives.

Keywords: Hepatoprotective, Phytoconstituents, Phytochemicals, Docking, *In silico*, *Ficus hispida*

INTRODUCTION

The liver, a pivotal organ in metabolic regulation and detoxification, is highly susceptible to a range of pathological conditions that can compromise its function, including viral hepatitis, alcohol-induced liver damage, and drug-induced toxicity. In recent years, there has been a growing interest in identifying natural products with hepatoprotective properties as potential therapeutic agents for liver disorders. Among the diverse range of medicinal plants used in traditional medicine, *Ficus hispida* L., commonly known as the Indian fig tree, has gained considerable attention for its potential hepatoprotective effects [1].

F. hispida, commonly known as the Indian fig tree or creeping fig, has garnered significant attention in traditional medicine for its diverse therapeutic properties. This plant, particularly its stem bark, is renowned for its pharmacological potentials, primarily due to its rich phytochemical profile, which includes flavonoids, saponins, tannins, and triterpenoids [2]. One of the most notable pharmacological activities of *F. hispida* is its hepatoprotective effect. Research has shown that extracts from the plant exhibit significant protection against liver damage caused by toxins or pathogens. This hepatoprotective action is attributed to the plant's ability to reduce liver enzyme levels and combat oxidative stress, thus preventing liver injury. In addition to its liver-protective properties, *F. hispida* demonstrates potent antioxidant activity. The plant's extracts have been proven to scavenge free radicals and enhance the body's antioxidant defenses [3]. This antioxidant capability is crucial for combating oxidative stress-related diseases, including cardiovascular disorders and cancer. Furthermore, *F. hispida* exhibits anti-inflammatory effects by inhibiting pro-inflammatory cytokines and enzymes. Its bioactive compounds effectively reduce inflammation in various experimental models, suggesting potential benefits for managing inflammatory conditions such as arthritis and ulcerative colitis [4]. The antimicrobial properties of *F. hispida* are also noteworthy. Studies have highlighted its effectiveness against a range of pathogens, including bacteria, fungi, and viruses, indicating its potential as a natural antimicrobial agent for treating infections [5]. In the realm of diabetes management, preliminary research suggests that *F. hispida* may possess antidiabetic properties, as its extracts have been observed to lower blood glucose levels and improve insulin sensitivity in diabetic models. Additionally, *F. hispida* has shown promise in cancer research. Extracts from the plant have been found to induce apoptosis, or programmed cell death, in cancer cells and inhibit tumor growth, underscoring its potential as a complementary therapeutic agent in cancer

treatment [6]. The plant is also traditionally used for wound healing, with its bioactive compounds contributing to enhanced repair and regeneration of wounds through their antimicrobial and anti-inflammatory properties. *F. hispida* stands out for its extensive pharmacological potentials, including hepatoprotection, antioxidant defense, anti-inflammatory action, antimicrobial activity, antidiabetic effects, anticancer properties, and wound healing. Continued research into these effects will further illuminate the plant's therapeutic potential and support its application in modern medicine [7].

Despite the promising traditional use and initial research findings, the detailed mechanisms by which these phytochemicals exert their hepatoprotective effects remain inadequately explored. In this context, *in silico* methods offer a valuable approach to predict and elucidate the interactions between phytochemicals and biological targets. Computational tools enable the virtual screening of phytochemicals against liver-specific molecular targets, providing insights into their potential efficacy and safety as hepatoprotective agents.

This research manuscript aims to conduct an *in silico* exploration of various phytochemicals (**Figure 1**) present in the stem bark of *F. hispida* to identify and evaluate their potential as hepatoprotective agents. By utilizing molecular docking techniques, this study seeks to elucidate the binding affinities of phytochemicals [2-demethoxytylophorine **(1)**, lupeol acetate **(2)**, *n*-triacontanol **(3)**, oleanolic acid **(4)**, hispidin **(5)**, terpeneol **(6)**, β-amyrine **(7)**, linalool **(8)**, gluanol **(9)**, β-sitosterol **(10)**, and bergaptine **(11)**] with key liver enzymes and receptors implicated in hepatoprotection. The results from this computational investigation are expected to provide a foundation for further experimental validation and therapeutic development of *F. hispida*-derived hepatoprotective agents. The integration of traditional knowledge with modern computational methods holds the promise of uncovering novel hepatoprotective compounds from *F. hispida*, thus contributing to the development of effective and natural therapeutic strategies for liver health.

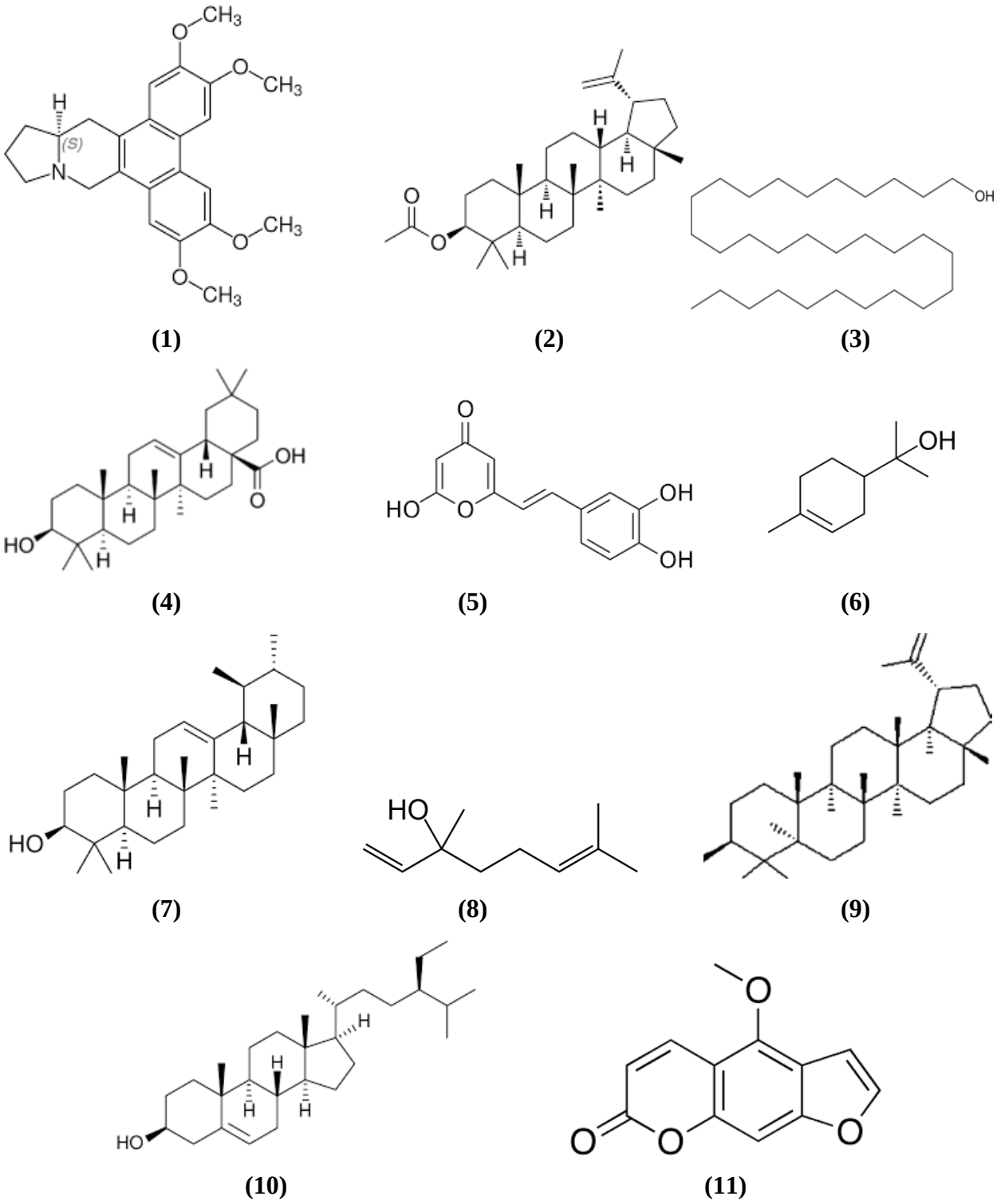

Figure 1. Structure of phytoconstituents.

MATERIALS AND METHODS

Preparation of Ligand

The 3D structure of chosen ligand was obtained in ".sdf" format using PubChem (https://pubchem.ncbi.nlm.nih.gov/). PubChem is an open-access database of chemical substances and biological activity. The method addressed the docking issue using flexible ligands and moveable protein atoms. The Avogadro programme was used to add hydrogen atoms to ligands, and the MMFF94 force field is utilized to compute the energy of the protein-ligand combination for every given configuration without any fitting parameters [8,9].

Preparation of Protein

3D crystalline target structures was downloaded from the Protein Data Bank (PDB), [PDB ID: 1IL6; Human Interleukin-6, NMR, Minimized Average Structure; https://doi.org/10.2210/pdb1IL6/pdb]. The target was created by removing all water molecules beyond 5A°, assigning disulfide links, bond order, and formal charges, and removing metal ions, co-factors, and heterogroup from the useable preprocessed and studied structure. With the assistance of the H-bond assignment technique, the hydrogen atoms as well as the hydrogen-bonding network were optimized. Molecular docking was used to estimate receptor grids for protein targets where the ligand would mix within the predicted active site. The grids (cubic boxes with defined dimensions) encompass the whole ligand and were built at the ligand's centroid (crystallized with the target structure). The grid box size was increased to 126 A°, 126 A° and 126 A° (x, y, and z, respectively) to include all of the amino acid residues present in stiff macromolecules. The Auto Grid 4.2, which came with Auto Dock 4.2, was used to generate grid maps. The grid points were 0.375° apart. The Van der Waals scale factor was set to 1.0, while the charge cutoff was set at 0.25. Induced-fit docking (IFD) was conducted on each ligand, and the lowest resulting score for the best-docked posture was confirmed [10,11].

Docking Procedure

The IFD was created utilizing the structure-based drug design technique, which involves rendering precise geometry ligands to dock with a biological target's defined structure. The free-state ligands are docked into the rigid state receptor's active site, enzyme, tube, etc., resulting in a predicted binding mode and the strength of the fit being evaluated. In receptor-based computational techniques, the attachment of a low-molecular-weight ligand to a macromolecular protein has its own significance since the most suitable connection with low energy values and possible steric conflicts is found. To investigate a particular docking issue, Auto Dock provides a

number of search methods. In this study, the Lamarckian Genetic Algorithm (LGA) was employed to identify the best conformers. During the docking process, a maximum of 10 conformers were evaluated. The population was limited to 150 individuals, who were selected at random. The mutation rate was set to 0.02 and the crossover rate was set to 0.8. The maximum number of energy evaluations was set to 500000, the maximum number of generations was set to 1000, the maximum number of top individuals that automatically survived was set to 1. Translations had a 0.2 step size, quaternions had a 5.0° step size, and torsions had a 5.0° step size. Cluster tolerance was set to 0.5, external grid energy to 1000.0, maximum binding energy to 0.0, maximum number of retries to 10000, and 10 LGA runs were performed. The interactions and binding energy of the docked structure were studied using the Auto Dock findings. It was performed many times to get different docked conformations as well as to assess anticipated docking energy. The optimal ligand-receptor structure was selected among the docked structures based on the ligand's lowest energy and minimum solvent accessibility. The Accelrys Visualizer discovery studio tool was used to visualize the docking findings [12,13].

RESULTS

The docking study revealed that compound named Bergaptine **(11)**, present in *F. hispida* demonstrated the highest inhibition of the hepatoprotective target Human Interleukin-6 with docking score of -7.71 Kcal/mol (**Table 1**) by interacting with amino acid residues ARG: 38, DA: 8 (H1), DT: 6 (H2), DT: 5 (H3), DT: 9 (H4), DT: 10, DC: 11, DA: 7 by forming 4 hydrogen bonds whereas the well-known compound β-Amyrine **(7)** presented the lowest inhibition of the hepatoprotective target Human Interleukin-6 with docking score of -4.85 Kcal/mol by interacting with amino acid residue LYS: 122 (H1) by forming 1 hydrogen bond (H1: Distance = 2.53 Å). Compounds such as Oleanolic acid (Dock Score: -6.02 Kcal/mol by interacting with TRP: 383 (H1), CYS: 419 (H2), ARG: 386 (H3), THR: 418 (H4), ASN: 415 (H5), ASP: 459, ARG: 457) **(4)**, Terpeneol **(6)**, and Linalool **(8)** showed moderate to good inhibitory activity against Human Interleukin-6 (**Figure 2**).

Few compounds like 2-demethoxytylophorine (Dock Score: -5.98 Kcal/mol by interacting with ASP: 492 (H1), GLU: 491 (H2)) **(1)**, β-Sitosterol (Dock Score: -5.79 Kcal/mol by interacting with ARG: 260 (H1), LEU (H2), VAL: 262, VAL: 265) **(10)**, Lupeol acetate (Dock Score: -5.61 Kcal/mol by interacting with GLY: 120 (H1), ASP: 149 (H2)) **(2)**, Hispidin

(Dock Score: -5.46 Kcal/mol by interacting with GLU: 667 (H1), GLU: 667 (H2), PRO: 674 (H3)) **(5)**, *n*-Triacontanol (Dock Score: -5.32 Kcal/mol) **(3)**, and Gluanol (Dock Score: -5.19 Kcal/mol by interacting with GLU: 667 (H1), GLU: 667 (H2), PRO: 674 (H3)) **(9)** demonstrated low to moderate inhibitory activity against Human Interleukin-6.

Table 1. Docking scores of phytoconstituents present in *Ficus hispida*.

S. No.	Compound Name	Binding Energy (kcal/mol)	No. of H Bonds	Interacting residue	Grid Point Spacing	Grid points	Coordinates of Central Grid Point of Map
1	2-Demethoxytylophorine	**-5.98**	02 (H1: Distance = 2.34 Å, H2: Distance = 2.15 Å)	ASP: 492 (H1), GLU: 491 (H2)	1.000 Angstroms	45 x, 65 y, 59 z	-37.126, -12.897, -68.374
2	Lupeol acetate	**-5.61**	02 (H1: Distance = 2.29 Å, H2: Distance = 2.34 Å)	GLY: 120 (H1), ASP: 149 (H2)	1.000 Angstroms	46 x, 50 y, 42 z	25.696, 26.392, 6.769
3	*n*-Triacontanol	**-5.32**	00	-	1.000 Angstroms	52 x, 55 y, 64 z	0.563, 0.284, -1.229

4	Oleanolic acid	**-6.02**	05 (H1: Distance = 2.4 Å, H2: Distance = 3.30 Å, H3: Distance = 1.86 Å, H4: Distance = 2.00 Å, H5: Distance = 1.83 Å)	TRP: 383 (H1), CYS: 419 (H2), ARG: 386 (H3), THR: 418 (H4), ASN: 415 (H5), ASP: 459, ARG: 457	1.000 Angstroms	66 x, 98 y, 110 z	106.129, -4.253, 18.577
5	Hispidin	**-5.46**	03 (H1: Distance = 1.83 Å, H2: Distance = 1.94 Å, H3: Distance = 2.93 Å	GLU: 667 (H1), GLU: 667 (H2), PRO: 674 (H3)	1.000 Angstroms	72 x, 64 y, 56 z	-47.126, -22.878, -68.384
6	Terpeneol	**-6.63**	00	-	1.000 Angstroms	50 x, 60 y, 58 z	6.274, 32.058, 72.983
7	β-Amyrine	**-4.85**	01 (H1: Distance = 2.53Å)	LYS: 122 (H1)	1.000 Angstroms	48 x, 48 y, 40 z	24.057, -26.699, -12.908
8	Linalool	**-6.42**	00	-	1.000 Angstroms	70 x, 68 y, 58 z	0.441, 0.195, -1.114

9	Gluanol	**-5.19**	03 (H1: Distance = 2.4 Å, H2: Distance = 3.30 Å, H3: Distance = 1.86 Å)	GLU: 667 (H1), GLU: 667 (H2), PRO: 674 (H3)	1.000 Angstroms	43 x, 50 y, 43 z	101.238, -5.364, 19.689
10	β-Sitosterol	**-5.79**	02 (H1: Distance = 2.34 Å, H2: Distance = 2.15 Å)	ARG: 260 (H1), LEU (H2), VAL: 262, VAL: 265	1.000 Angstroms	58 x, 60 y, 58 z	-36.014, -11.986, -57.263
11	Bergaptine	**-7.71**	04 (H1: Distance = 1.83 Å, H2: Distance = 1.94 Å, H3: Distance = 2.93 Å, H4: Distance = 2.43 Å)	ARG: 38, DA: 8 (H1), DT: 6 (H2), DT: 5 (H3), DT: 9 (H4), DT: 10, DC: 11, DA: 7	1.000 Angstroms	40 x, 46 y, 40 z	24.584, 25.281, 5.658

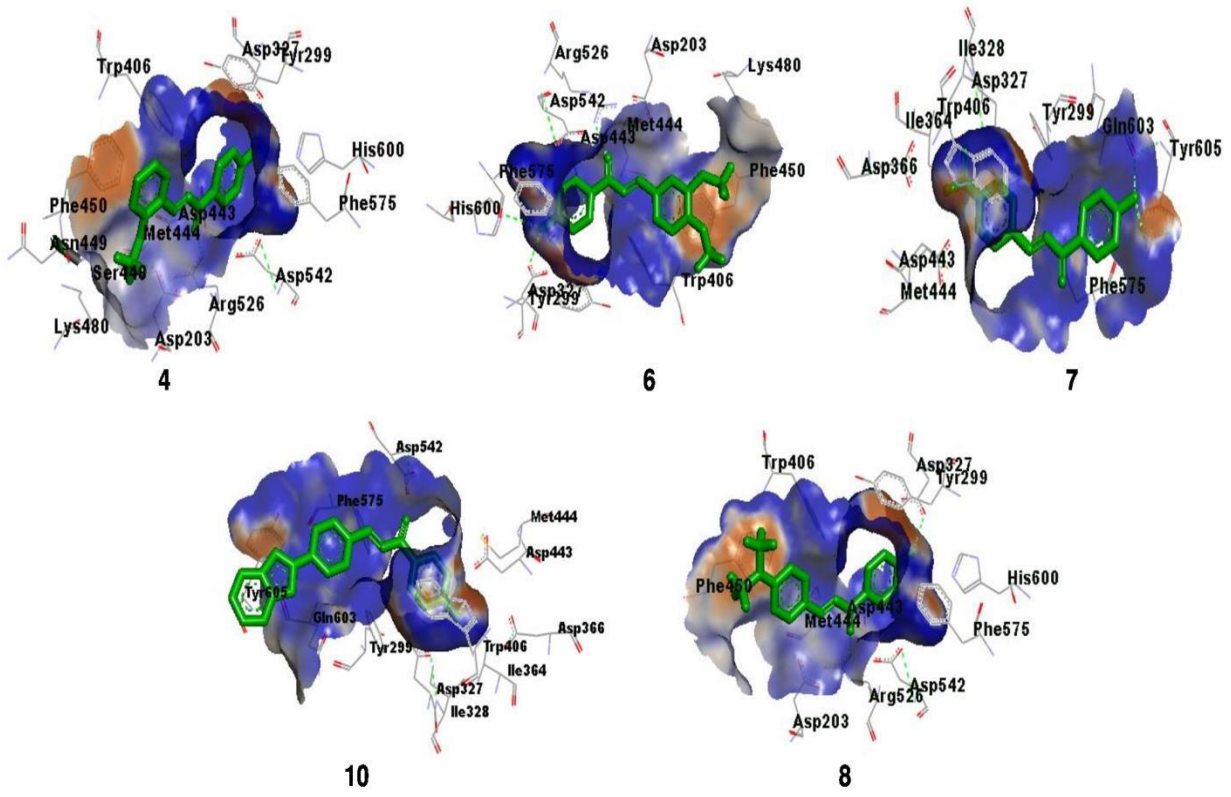

Figure 2. Docking poses of Top-5 compounds against Human Interleukin-6.

DISCUSSION

The hepatoprotective action of the medicines was confirmed and explained by the induced-fit molecular docking studies. For a long time, this method was the go-to for figuring out how chemicals interact with proteins and where ligands attach to specific targets. A model of the binding affinity of the inhibitors was constructed using the molecular docking approach [14]. Docking score, hydrogen bonds, Van der Waal's interactions, π-cation interaction, and π-π stacking interactions were the primary factors evaluated for the outcome prediction. We used these factors to determine the protein ligand affinity. A negative docking score indicates a positive binding affinity. The results of the docking process indicate the presence of many important characteristics: Antagonistic ligands have a high binding affinity for the molecular target and increased Van der Waals interactions, whereas ligands with multiple bulky groups have extra hydrogen bonding connections. Accordingly, the IFD module allows for the prediction of inhibitor binding [15].

It was easy to see that hydrophobic amino acid residues encircled both molecules. Due to their inability to evaporate into the surrounding solvent, it follows that neither chemical could access the protein's active site binding cleft. Because of their diminutive size, the ligands may be able to reach more remote areas of the cavity. Applying surface representation rendered the ligand invisible at the cavity owing to its utter obliteration by amino acid residues. When ligands are snugly packed together, they generate very stable Van der Waals interactions. By avoiding solvent exposure, chemicals were able to reach the protein's active site. The ligand is able to form strong connections in the target's active site because amino acid residues form a deep bent-shaped cleft. The amino function may have a major role in the enhanced target inhibition, according to one theory [16]. The location of interacting oxygen groups is crucial, according to the research. As this hypothesis goes, it might be possible to enhance contact with the active site by positioning the hydroxyl group farthest from the scaffold and the keto group closest it. The stability of the drug-target relationship may be influenced by steric, hydrophobic, electrostatic, and hydrophilic interactions, among others. The associations were confirmed by molecular docking studies, which accurately predicted the drugs' theoretical binding to the biological target [17].

CONCLUSION

The *in silico* exploration of various phytochemicals present in the stem bark of *F. hispida* has provided compelling evidence supporting its potential as a hepatoprotective agent. The findings from molecular docking studies suggest that several bioactive compounds within the plant exhibit strong binding affinities to key proteins involved in liver function and detoxification pathways, indicating their possible role in mitigating liver damage and enhancing hepatic health. This study reinforces the traditional use of *F. hispida* in liver-related ailments and opens new avenues for further experimental validation and development of novel hepatoprotective therapies derived from this plant. The results not only underscore the therapeutic potential of *F. hispida* but also highlight the value of *in silico* methods in the early-stage screening of natural compounds for drug discovery. Future research should focus on experimental studies to confirm these findings and explore the clinical applicability of these phytochemicals in the prevention and treatment of liver diseases.

Conflict of Interest

Declared none

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Animal ethical permission

Not required

REFERENCES

1. Bairwa R, Sharma P, Khandelwal S. Pharmacognostic and phytochemical study of *Ficus hispida* Linn. (Moraceae). Int J Pharm Sci Res. 2012;3(9):3130-4.
2. Kumar P, Shukla A. Pharmacological profile of *Ficus hispida*: A review. J Ethnopharmacol. 2015;168:99-106.
3. Kumar V, Sharma P. Evaluation of the anti-diabetic activity of *Ficus hispida* fruit extract in diabetic rats. Int J Green Pharm. 2014;8(3):220-4.
4. Mohan P, Shukla P. Isolation and characterization of bioactive compounds from *Ficus hispida* Linn. leaves and their antimicrobial activity. Asian J Pharm Clin Res. 2016;9(5):142-6.
5. Singh S, Singh M. Traditional uses and pharmacological activities of *Ficus hispida*: A comprehensive review. Pharmacol Rev. 2017;11(21):106-14.
6. Gautam S, Mishra P. Phytochemical and antimicrobial screening of *Ficus hispida* leaves. J Chem Pharm Res. 2018;10(3):20-4.
7. Mishra S, Singh R. Evaluation of antioxidant and anti-inflammatory potential of *Ficus hispida* extracts. J Food Sci Technol. 2019;56(6):2952-61.
8. Asati V, Bajaj S, Mahapatra DK, Bharti SK. Molecular modeling studies of some thiazolidine-2, 4-dione derivatives as 15-PGDH inhibitors. Med Chem Res. 2016;25:94-108.

9. Chhajed SS, Chaskar S, Kshirsagar SK, Haldar GA, Mahapatra DK. Rational design and synthesis of some PPAR-γ agonists: Substituted benzylideneamino-benzylidene-thiazolidine-2, 4-diones. Comp Biol Chem. 2017;67:260-5.

10. Joseph TM, Suresh AM, Mahapatra DK, Haponiuk J, Thomas S. The Efficacious Benefit of 25-Hydroxy Vitamin D to Prevent COVID-19: An In-Silico Study Targeting SARS-CoV-2 Spike Protein. Nutrients. 2022;14(23):4964.

11. Asati V, Bharti SK, Rathore A, Mahapatra DK. SWFB and GA strategies for variable selection in QSAR studies for the validation of thiazolidine-2, 4-dione derivatives as promising antitumor candidates. Indian J Pharm Educ Res. 2017;51(3):436-51.

12. Pandey R, Dubey I, Ahmad I, Mahapatra DK, Patel H, Kumar P. In-Silico Study of Some Dexamethasone Analogs and Derivatives against SARS-CoV-2 Target: A Cost-Effective Alternative to Remdesivir for Various COVID Phases. Current Chinese Science (Bioinformatics). 2022;2(4):294-309.

13. Singh RK, Mishra AK, Kumar P, Mahapatra DK. Molecular Docking and In Vivo Screening of Some Bioactive Phenoxyacetanilide Derivatives as Potent Non-Steroidal Anti-Inflammatory Drugs. Int J Cur Res Rev. 2021;13(10):189.

14. Zehra A, Nisha R, Kumar A, Nandan D, Ahmad I, Mahapatra DK, Patel H, Maity B, Kumar P. Computational Approaches Molecular Docking and MD Simulation Establishes the Potential COVID-19 Main Protease Inhibitors from Natural Products. Current Chinese Science (Bioinformatics). 2024;4(2):114-34.

15. Joseph TM, Mahapatra DK. Exploration of two natural inhibitors for treating COVID-19 from simple natural products: nature probably has all the Solutions. Journal of Public Health and Sanitation. 2020;1(1):17-24.

16. M Dadure K, GM Haldar A, Kar Mahapatra D. Molecular Docking Study of Substituted Chalcone Compounds as Potential Cyclin-Dependent Protein Kinase 2 (Cdk-2) Inhibitors. Advanced Materials Proceedings. 2020;5(4):1-3.

17. Mahapatra DK, Dadure KM, Haldar AG. In silico Computation of Theoretical Physicochemical Properties and Pharmacokinetic Profiles of Some Novel Substituted Amodiaquine Based Compounds. International Journal of Chem-informatics Research. 2020;6(2):9-16.