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MOLECULAR DOCKING AND ADMET ANALYSIS OF THE LEUCAS ASPERA PLANT CONSTITUENT WITH MULTIPLE PHARMACOLOGICAL ACTIVITIES

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ABSTRACT

Leucasaspera belongs to the Lamiaceae family and stands out for its pharmacological and therapeutic traits—packed full of active biological compounds. Finding new lead molecules from natural sources drives progress in modern drug discovery. In this study, I used a two-step computational approach: first, in silico method of molecular docking, then another one is ADMET analysis. This let me systematically evaluate the therapeutic potential of major phytochemicals isolated from Leucasaspera by focusing on phytochemicals like flavonoids, alkaloid, phytosterol, terpenoids and terpene derivatives, and phenolic acids. Each was screened against 16 well-established protein targets that play key roles in eight major therapeutic areas: anti-rheumatoid, anti-asthma, anti-inflammatory, anti-malaria, appetite stimulant, anti-emetic, anti-migraine and anti-flu activities. Binding free energy measured molecular affinity, while drug-likeness rules and predictive toxicity models gave insight into pharmacokinetic properties and safety. Phytosteroids such as sitosterol and campesterol showed high binding affinity, thus indicates strong interactions between drug and protein. The docking results showed that several phytochemicals from leucasaspera have strong affinities for multiple protein targets. This points to their multi-functional therapeutic potential. Overall, this study of molecular docking and ADMET analysis highlights leucasaspera phytochemicals has potential for drug development.

Key Words: Leucasaspera, molecular docking, ADMET analysis, phytosteroids, baicalein, anti-rheumatoid, anti-inflammatory, anti-asthma, anti-emetic, anti-migraine, anti-flu, appetite stimulant, anti-malaria.

INTRODUCTION

Drug discovery is a fundamental area of pharmaceutical sciences focused on identifying and developing chemical substances that produce therapeutic effects with acceptable safety and efficacy. It is not only about synthetic chemistry but also includes natural product research, pharmacology, medicinal chemistry, pharmaceutical analysis, and computational approaches. Medicinal plants have historically been a major source of drugs, supplying bioactive compounds which produce therapeutic activity that form the basis of both traditional and modern medicine.

Traditional medicinal systems have long used plants to treat conditions such as inflammation, infections, gastric disorders, respiratory diseases, metabolic disorders, neurological conditions, and mental health.

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Issues the plant may use entirely whole plant or may parts of plants like leaves, fruit, roots, latex or also used in various forms like dried powdered, or fresh paste or may in extract which obtained with usage of various solvents and methods. Their therapeutic effects arise from phytochemicals such as alkaloids, flavonoids, terpenoids, glycosides, phenolics, tannins, saponins, and steroids, many of which show clinically proven pharmacological activities.

Scientific validation of herbal drugs requires proper identification, structural characterization, and quality control. Instrumental Methods of Analysis play a key role in this process through techniques such as UV-Visible spectroscopy, IR spectroscopy, HPLC, GC-MS, LC-MS, and NMR, which help determine the structure, purity, and composition of phytoconstituents.

However, analytical techniques alone cannot explain how these compounds exert biological effects. For that purpose, the in silico method can be used which is computer-based simulations, modeling, and analyses used

to study biological, chemical, and physical processes. Molecular docking complements these methods by predicting the interaction between phytochemicals and biological targets such as enzymes and receptors. It provides details about binding affinity, inhibition constant, interaction patterns, interaction residues and mechanisms of action while reducing experimental cost and time. Also provides an solid base for further tests and trials.

Leucasaspera is an erect, aromatic annual herb growing up to 60 cm in height. Stem: Quadrangular, hairy, and branched, Leaves: Opposite, linear to lanceolate, pubescent, with serrated margins, Flowers: White, sessile, arranged in dense axillary whorls, Calyx: Tubular, hairy, with stiff bristles, Corolla: Bilabiate, typical of Lamiaceae, Fruit: Nutlets enclosed within persistent calyx. These morphological features aid in botanical identification and pharmacognosticevaluation. Leucasaspera is widely distributed across India, Sri Lanka, Nepal, Bangladesh, and Southeast Asia. It commonly grows in dry grasslands, roadsides, agricultural fields, and wastelands. The plant adapts well to tropical and subtropical climates and flourishes during the monsoon season, exhibiting high resilience to drought and poor soil conditions.

In Ayurveda, Siddha, and Unani medicine, Leucasaspera is considered a medicinal weed with broad therapeutic applications. Traditionally, it is used for: Treatment of fever, cold, cough, and asthma, Relief from inflammatory conditions and pain. Management of skin diseases and wound healing. Treatment of snake bites, scorpion stings, and insect bites. Alleviation of headache and ear infections.

MATERIALS AND METHODS

Software Used

- Discovery Studio Visualizer – visualize & clean protein
- Autodock4 – to perform molecular docking

- ChemSketch/ChemDraw – draw ligand structure
- Online SMILES translator – convert structure → .pdb
- MGL Tools (AutoDock&AutoGrid) – docking process
- Cygwin terminal – run docking commands

Selection of Medicinal Plant

The medicinal plant Leucasaspera (Willd.) Link was selected for the present study based on its extensive use in traditional medicine and reported pharmacological activities.

Collection of Phytochemical Data

Reported phytochemical constituents of Leucasaspera were collected from published research articles, phytochemical databases, and review literature which is collective available in IMPPAT (Indian Medicinal Plants, Phytochemistry and Therapeutics) database. Major compounds selected for docking studies included:

Selection of Target Protein

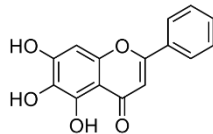
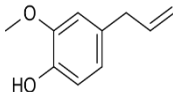
Based on the selected pharmacological activities, the two target proteins are selected for each pharmacological activity. The selection of protein by availability of structure of specific protein of specific organisms in RCSB protein database. The protein should possess,

- Availability of high-resolution crystal structure
- Relevance to disease condition
- Presence of a well-defined active site

Ligand Preparation

In order to retrieve 3D structure of phytochemicals by, copying the smiles from the pubchem database then paste it on cactus Online SMILES Translator web tool by NCI/CADD Group and downloaded in required format. Open in AutoDock Tools, Add Gasteiger charges then proceed torsions (rotatable bonds) and Save as PDBQT (flexible ligand)

Table 1: Structure of Phytochemical constituents

Sl. No.	Chemical class	Chemical constituents	Structure
1	Flavonoid	Baicalein	
2	Phenolic compound	Eugenol	

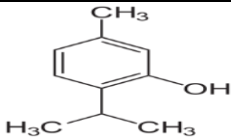
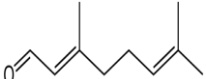
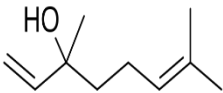
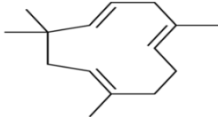
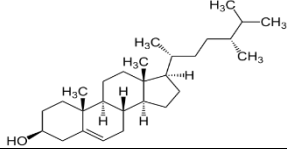
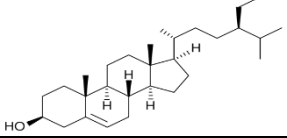
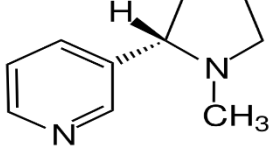
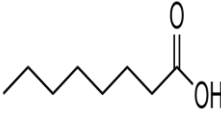
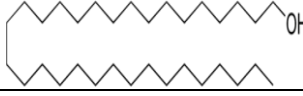
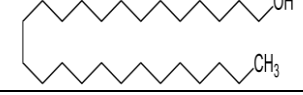
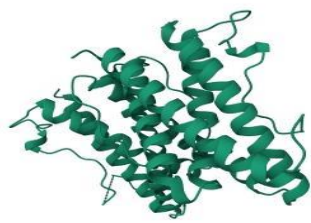
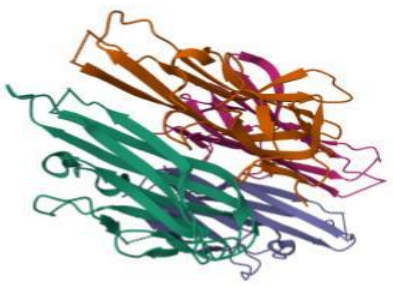
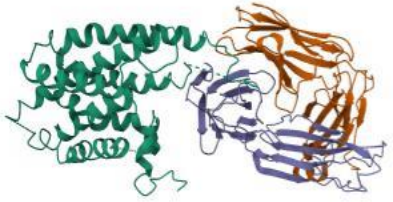
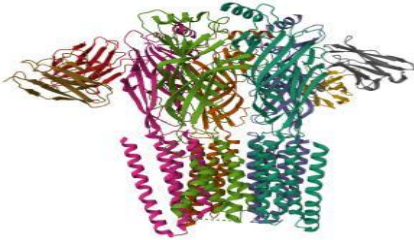
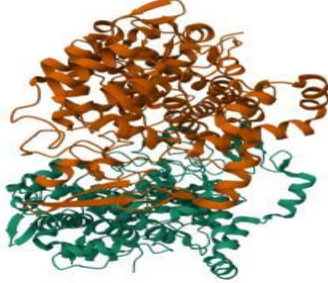
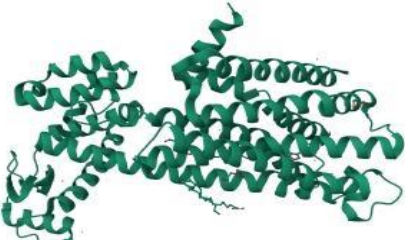
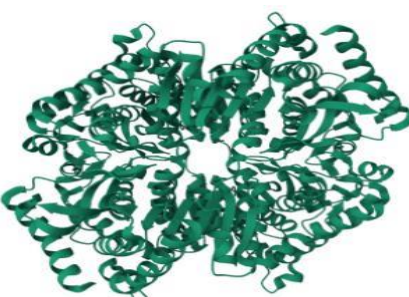
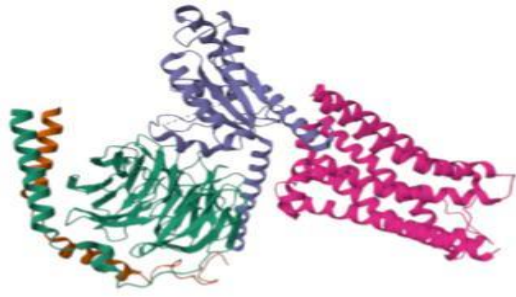
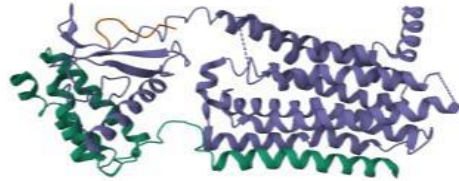
3	Terpenoids and Terpene Derivatives	Thymol	
		Citral	
		Linalool	
		Humulene	
4	Phytosterol	Campesterol	
		β -Sitosterol	
5	Alkaloid	Nicotine	
6	Fatty acid	Octanoic acid	
		Stearic acid	
7	Long-chain aliphatic alcohol	Dotriacontanol	
		Hexacosanol	
8	Aliphatic hydrocarbon	Decane	
		Hexadecane	

Table 2: Structure of Target Protein

1	Anti-rheumatoid	Interleukin-6	1ALU	
		TNF-alpha	2AZ5	
2	Anti-asthma	B2-adrenergic receptor	2R4R	
		Phosphodiesterase	1WOJ	

3	Appetite stimulant	Alpha-2 adrenergic receptor	6KUX	
		GABA-A	6D6T	
4	Anti-inflammatory	COX-1	6Y3C	
		COX-2	5IKR	
5	Anti-emetic	NK (tachykinin)	6E59	

		Dopamine D2 receptor	6CM4	
6	Anti-malaria	Falcipain-2	3BPF	
		Plasmodium lactate	1T24	
7	Anti-migraine	5-HT1b	6G79	
		Calcitonin gene related peptide	7KNU	

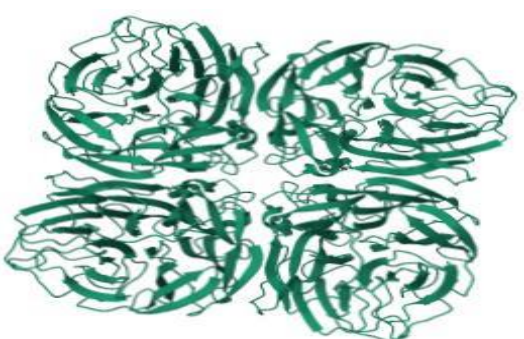
8	Anti-flu	Neuraminidase	1NNC	
		PA-endonuclease	5WA7	

Table 3: Docking scores for anti-Rheumatoid activity

Sl. No.	Ligand name (phytochemicals)	Protein-1 ID :	1ALU	Protein-2 ID :	2AZ5
		Binding Energy	Inhibition Constant	Binding energy	Inhibition Constant
a	Baicalein	-6.66	13.14 μ M	-6.69	12.40 μ M
b	Campesterol	-7.42	3.64 μ M	-9.58	94.41 nM
c	Citral	-5.07	190.84 μ M	-4.92	246.39 μ M
d	Decane	-3.87	1.45 mM	-4.19	852.63 μ M
e	Dotriacontanol	-3.96	1.25 mM	-4.04	1.09 mM
f	Nicotine	-4.50	505.55 μ M	-4.70	359.74 μ M
g	Hexadecane	-4.72	345.83 μ M	-4.76	324.77 μ M
h	Stearic acid	-5.14	171.50 μ M	-5.13	174.80 μ M
i	Octanoic acid	-5.28	135.11 μ M	-5.21	150.84 μ M
j	Hexacosanol	-2.63	11.73 mM	-4.15	900.40 μ M
k	Eugenol	-5.57	82.36 μ M	-4.54	468.42 μ M
l	Humulene	-5.75	61.15 μ M	-6.85	9.57 μ M
m	Thymol	-4.81	295.92 μ M	-5.21	150.64 μ M
n	Linalool	-4.35	646.67 μ M	-5.18	158.62 μ M
o	Sitosterol	-7.61	2.64 μ M	-9.96	50.11 nM

Table 4: Docking scores for anti-asthma activity

Sl. No.	Ligand name (phytochemicals)	Protein-1 ID :	2R4R	Protein-2 ID :	1WOJ
		Binding Energy	Inhibition Constant	Binding energy	Inhibition Constant
a	Baicalein	- 7.71	2.21 μ M	-7.02	7.16 μ M
b	Campesterol	- 9.43	122.97 nM	-9.18	186.76 μ M
c	Citral	- 5.29	132.41 μ M	-4.72	344.89 μ M
d	Decane	- 4.69	367.77 μ M	-4.58	437.65 μ M
e	Dotriacontanol	- 3.03	6.02 nM	-3.80	1.63mM
f	Nicotine	- 5.61	76.59 μ M	-4.66	383.43 μ M
g	Hexadecane	- 5.80	55.97 μ M	-4.41	581.11 μ M

h	Stearic acid	- 6.92	8.51 μ M	-4.55	458.30 μ M
i	Octanoic acid	- 5.13	173.41 μ M	-4.86	273.85 μ M
j	Hexacosanol	- 3.96	1.25 mM	-3.91	1.37 mM
k	Eugenol	- 4.75	327.91 μ M	-5.50	92.99 μ M
l	Humulene	- 6.83	9.82 μ M	-6.65	13.44 μ M
m	Thymol	- 5.21	152.05 μ M	-5.48	96.20 μ M
n	Linalool	-5.33	124.08 μ M	-4.77	319.69 μ M
o	Sitosterol	-10.43	22.72 nM	-9.17	190.89 μ M

Table 5: Docking scores for Appetite stimulant activity

Sl. No.	Ligand name (phytochemicals)	Protein-1 ID:	6KUX	Protein-2 ID:	6D6T
		Binding Energy	Inhibition Constant	Binding energy	Inhibition Constant
a	Baicalein	-7.04	6.89 μ M	-6.81	10.13 μ M
b	Campesterol	-9.21	176.64 nM	-8.11	1.13 μ M
c	Citral	-4.57	443.34 μ M	-5.19	157.61 μ M
d	Decane	-3.90	1.38 mM	-3.97	1.24mM
e	Dotriacontanol	-4.62	408.07 μ M	-1.80	48.09 mM
f	Nicotine	-4.67	377.67 μ M	-5.30	129.79 μ M
g	Hexadecane	-4.55	461.54 μ M	-4.35	650.15 μ M
h	Stearic acid	-5.16	165.35 μ M	-4.53	481.85 μ M
i	Octanoic acid	-5.04	202.17 μ M	-5.00	216.34 μ M
j	Hexacosanol	-3.87	1.47 mM	-2.24	22.90 mM
k	Eugenol	-4.32	679.01 μ M	-5.38	114.11 μ M
l	Humulene	-6.35	22.26 μ M	-6.25	26.23 μ M
m	Thymol	-4.96	232.31 μ M	-5.29	132.42 μ M
n	Linalool	-4.65	393.08 μ M	-4.87	271.36 μ M
o	Sitosterol	-10.30	28.41 μ M	-8.21	960.31 nM

Table 6: Docking scores for anti-inflammatory activity

Sl. No.	Ligand name (phytochemicals)	Protein-1 ID:	6Y3C	Protein-2 ID:	51KR
		Binding Energy	Inhibition Constant	Binding energy	Inhibition Constant
a	Baicalein	-8.24	911.69 nM	-7.52	3.09 μ M
b	Campesterol	-10.91	9.99 nM	-11.64	2.92 nM
c	Citral	-5.64	73.29 μ M	-5.79	57.30 μ M
d	Decane	-5.57	82.49 μ M	-5.37	116.69 μ M
e	Dotriacontanol	-5.06	196.0 μ M	- 5.69	67.44 μ M
f	Nicotine	-6.15	31.03 μ M	-5.50	93.26 μ M
g	Hexadecane	-6.08	34.66 μ M	-4.99	220.45 μ M
h	Stearic acid	-6.26	25.5 μ M	-5.78	57.72 μ M
i	Octanoic acid	-5.00	217.41 μ M	-5.98	41.32 μ M
j	Hexacosanol	-5.69	67.28 μ M	-5.82	54.42 μ M
k	Eugenol	-5.83	52.94 μ M	-6.69	12.47 μ M
l	Humulene	-8.27	872.35 μ M	-6.86	9.43 μ M
m	Thymol	-6.10	33.89 μ M	-6.62	14.05 μ M
n	Linalool	-6.13	31.91 μ M	-5.45	101.44 μ M
o	Sitosterol	-11.37	4.63 μ M	-11.49	3.80 nM

Table 7: Docking scores for Anti-emetic activity

Sl. No.	Ligand name (phytochemicals)	Protein-1 ID:	6E59	Protein-2 ID:	6CM4
		Binding Energy	Inhibition Constant	Binding energy	Inhibition Constant
a	Baicalein	-7.45	3.43 μ M	-6.47	18.20 μ M

b	Campesterol	-8.02	1.33 μ M	-10.68	14.97 nM
c	Citral	-4.57	448.13 μ M	-6.05	36.81 μ M
d	Decane	-4.77	320.10 μ M	-5.45	100.70 μ M
e	Dotriacontanol	-3.99	1.19 mM	-4.65	390.55 μ M
f	Nicotine	-5.47	98.65 μ M	-6.01	39.19 μ M
g	Hexadecane	-4.86	274.23 μ M	-5.99	40.96 μ M
h	Stearic acid	-4.71	352.78 μ M	-5.73	63.59 μ M
i	Octanoic acid	-4.27	739.67 μ M	-4.62	409.80 μ M
j	Hexacosanol	-5.10	181.32 μ M	-6.34	22.66 μ M
k	Eugenol	-4.45	544.67 μ M	-5.77	59.12 μ M
l	Humulene	-7.78	1.99 μ M	-6.24	26.72 μ M
m	Thymol	-4.99	219.54 μ M	- 6.78	10.74 μ M
n	Linalool	-4.55	458.36 μ M	-6.29	24.67 μ M
o	Sitosterol	-9.28	156.77 nM	-9.62	88.39 nM

Table 8: Docking scores for anti-malaria activity

Sl. No.	Ligand name (phytochemicals)	Protein-1 ID:	3BPF	Protein-2 ID:	1T24
		Binding Energy	Inhibition Constant	Binding energy	Inhibition Constant
a	Baicalein	-6.34	22.57 μ M	-7.65	2.48 μ M
b	Campesterol	-9.87	57.98 nM	-7.26	4.73 μ M
c	Citral	-5.22	149.49 μ M	-5.60	79.19 μ M
d	Decane	-5.46	99.68 μ M	-4.23	787.77 μ M
e	Dotriacontanol	-3.71	1.92 mM	-2.34	19.12 μ M
f	Nicotine	-5.66	71.56 μ M	-6.18	29.33 μ M
g	Hexadecane	-4.07	1.04 mM	-4.98	221.98 μ M
h	Stearic acid	-3.75	1.79 mM	-7.10	6.30 μ M
i	Octanoic acid	-3.53	2.58 mM	-5.95	43.74 μ M
j	Hexacosanol	-3.67	2.05 mM	-1.85	44.22 mM
k	Eugenol	-4.52	486.51 μ M	-5.40	109.21 μ M
l	Humulene	-6.12	32.74 μ M	-6.91	8.64 μ M
m	Thymol	-6.82	9.97 μ M	-5.89	48.46 μ M
n	Linalool	-5.72	64.05 μ M	-5.22	150.16 μ M
o	Sitosterol	- 8.92	287.95 μ M	-8.55	539.00 μ M

Table 9: Docking scores for anti-migraine activity

Sl. No.	Ligand name (phytochemicals)	Protein-1 ID:	6G79	Protein-2 ID:	7KNU
		Binding Energy	Inhibition Constant	Binding energy	Inhibition Constant
a	Baicalein	-7.37	3.95 μ M	-6.47	17.97 μ M
b	Campesterol	-7.42	3.62 μ M	-7.82	1.84 μ M
c	Citral	- 5.46	99.99 μ M	-5.21	150.80 μ M
d	Decane	-4.61	416.83 μ M	-4.16	885.32 μ M
e	Dotriacontanol	-2.03	32.46 mM	-2.46	15.85 mM
f	Nicotine	-6.23	27.08 μ M	-5.36	116.98 μ M
g	Hexadecane	-5.42	106.19 μ M	-4.21	826.73 μ M
h	Stearic acid	-4.52	482.19 μ M	-3.72	1.88 mM
i	Octanoic acid	-4.05	1.07 mM	-3.99	1.18 mM
j	Hexacosanol	-3.87	1.45 mM	-3.67	2.03 mM
k	Eugenol	-5.24	143.27 μ M	-5.21	152.62 μ M
l	Humulene	-7.37	3.96 μ M	-6.28	24.99 μ M
m	Thymol	-5.63	75.11 μ M	-5.57	83.32 μ M
n	Linalool	-5.35	120.23 μ M	-4.74	337.94 μ M
o	Sitosterol	-8.09	1.18 μ M	-8.21	964.18 nM

Table 10: Docking scores for anti-flu activity

Sl. No.	Ligand name (phytochemicals)	Protein-1 ID:	1NNC	Protein-2 ID:	5WA7
		Binding Energy	Inhibition Constant	Binding energy	Inhibition Constant
a	Baicalein	-5.55	85.20 μ M	-6.69	12.40 μ M
b	Campesterol	-7.64	2.50 μ M	-9.58	94.41 nM
c	Citral	-4.79	307.22 μ M	-4.92	246.39 μ M
d	Decane	-4.82	291.17 μ M	-4.19	852.63 μ M
e	Dotriacontanol	-0.84	243.83 mM	-4.04	1.09 mM
f	Nicotine	-5.48	96.88 μ M	-4.70	359.74 μ M
g	Hexadecane	-3.67	2.03 mM	-4.76	324.77 μ M
h	Stearic acid	-6.22	27.45 μ M	-5.13	174.80 μ M
i	Octanoic acid	-5.14	171.28 μ M	-5.21	150.84 μ M
j	Hexacosanol	-2.17	25.84 mM	-4.15	900.40 μ M
k	Eugenol	-4.22	803.27 μ M	-4.54	468.42 μ M
l	Humulene	-5.90	47.23 μ M	-6.85	9.57 μ M
m	Thymol	-4.54	472.70 μ M	-5.21	150.0 μ M
n	Linalool	-4.17	818.06 μ M	-5.18	158.62 μ M
o	Sitosterol	-7.79	1.94 μ M	-9.96	50.11 nM

Table 11: ADME studies of the phytochemical constituents

Phytochemicals	Molecular weight (g/mol)	HBA	HBD	Log p	GI absorption	BBB permeability	Skin permeability (cm/s)	Bio availability	Lipinski rule
Baicalein	270.24	5	3	2.24	High	No	-5.2	0.55	Yes
Campesterol	400.68	1	1	6.92	Low	No	-2.5	0.55	Yes
Citral	302.45	2	0	5.00	High	Yes	-4.2	0.55	Yes
Decane	142.28	0	0	4.16	Low	Yes	-3.61	0.55	Yes
Dotriacontanol	466.87	1	1	11.23	Low	No	2.06	0.55	Yes
Nicotine	162.23	2	0	1.48	High	Yes	-6.46	0.55	Yes
Hexadecane	226.44	0	0	6.42	Low	No	-1.80	0.55	Yes
Stearic acid	284.48	2	1	5.93	High	No	-2.19	0.85	Yes
Octanoic acid	144.21	2	1	2.22	High	Yes	-5.01	0.85	Yes
Hexacosanol	382.71	1	1	9.06	Low	No	0.26	0.55	Yes
Eugenol	164.20	2	1	2.25	High	Yes	-5.69	0.55	Yes
Humulene	204.35	0	0	4.26	Low	No	-4.32	0.55	Yes
Thymol	150.22	1	1	2.80	High	Yes	-4.88	0.55	Yes
Linalool	154.25	1	1	2.66	High	Yes	-5.13	0.55	Yes
Sitosterol	414.71	1	1	7.24	Low	No	-2.20	0.55	Yes

Table 12: Toxicity studies of the phytochemical constituents

Phytochemicals	Total clearance (ml/min/kg)	AMES toxicity	Max. tolerated dose (mg/kg/day)	Hepatic toxicity	Skin sensitization	Minnow toxicity (log mm)
Baicalein	0.252	No	0.498	No	No	1.25
Campesterol	0.572	No	-0.641	No	No	-1.807
Citral	0.376	No	0.543	No	Yes	0.945
Decane	1.615	No	0.463	No	No	0.154
Dotriacontanol	2.245	No	-0.284	No	Yes	-4.782
Nicotine	0.86	No	0.62	Yes	Yes	1.777
Hexadecane	1.85	No	0.141	No	Yes	-1.409
Stearic acid	1.832	No	-0.791	No	Yes	-1.565
Octanoic acid	1.693	Yes	0.914	No	No	4.299
Hexacosanol	2.104	No	-0.396	No	Yes	-3.337

Eugenol	0.282	Yes	1.024	No	Yes	1.702
Humulene	1.282	No	0.551	No	Yes	0.716
Thymol	0.211	No	1.007	Yes	Yes	1.213
Linalool	0.446	No	0.771	No	Yes	1.306
Sitosterol	0.628	No	-0.621	No	No	-1.802

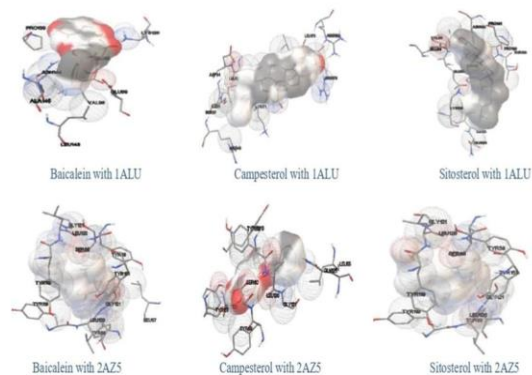


Figure 2: Docking visuals of anti-Rheumatoid activity

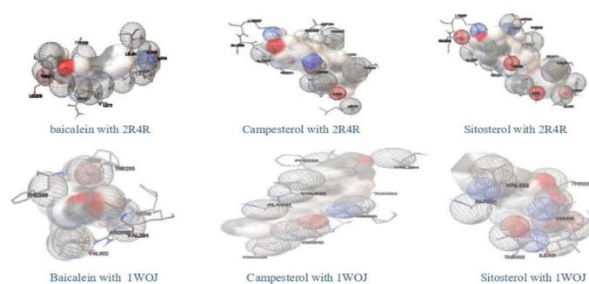


Figure 3: Docking visuals of anti-asthma activity

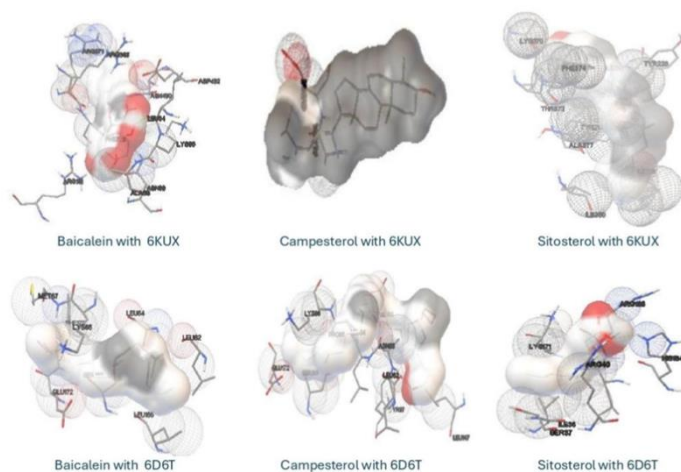


Figure 4: Docking visuals of Appetite stimulant activity

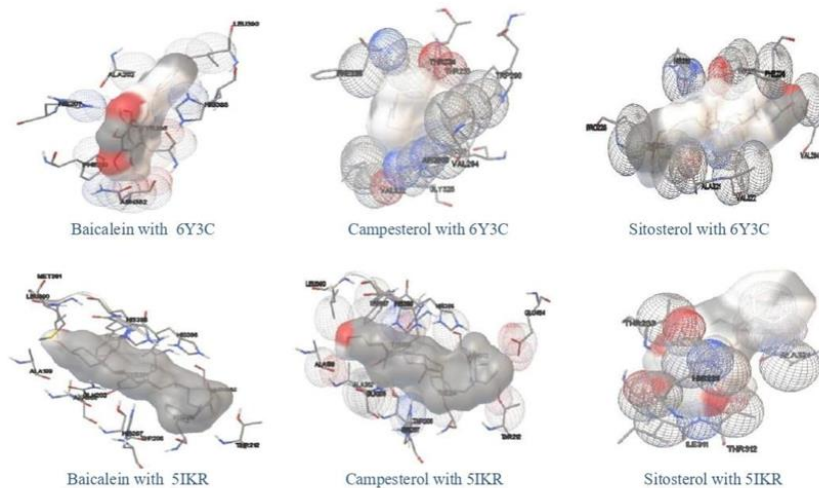


Figure 5: Docking visuals of Anti-inflammatory activity

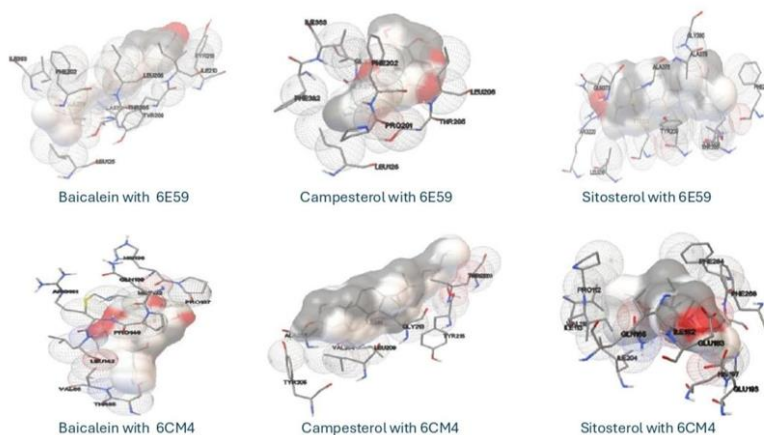


Figure 6: Docking visuals of Anti-emetic activity

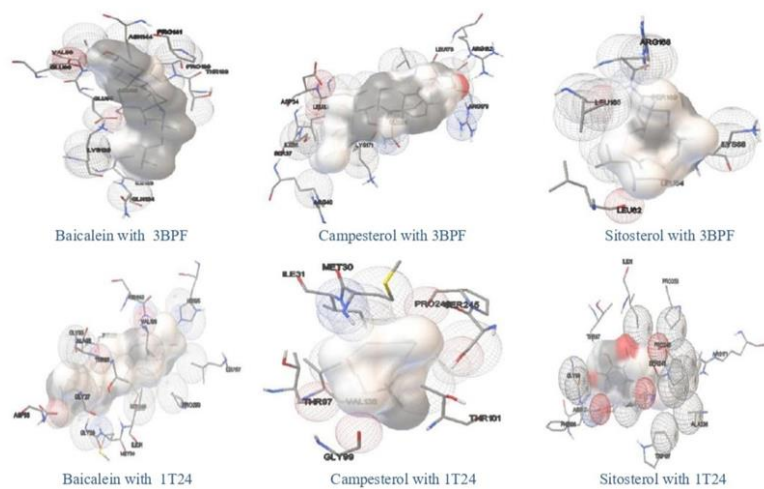


Figure 7: Docking visuals of Anti-malarial activity

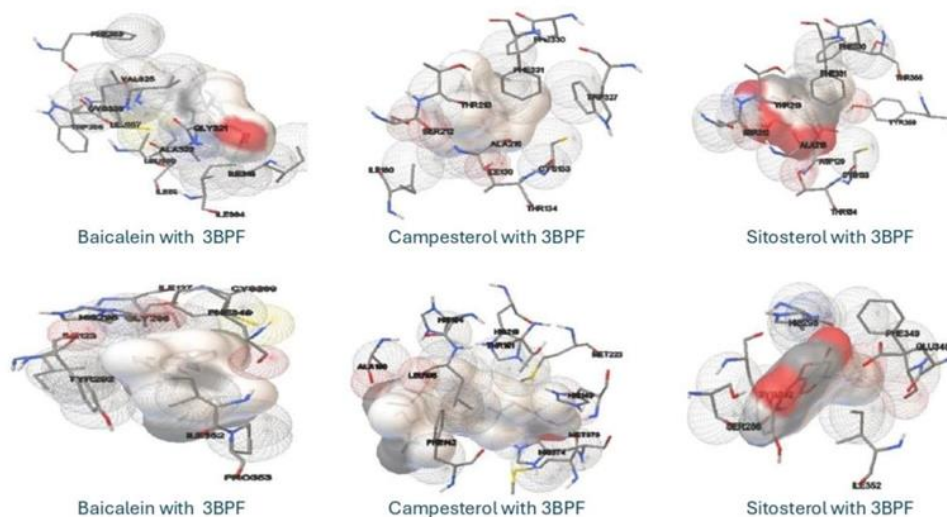


Figure 8: Docking visuals of Anti-migraine activity

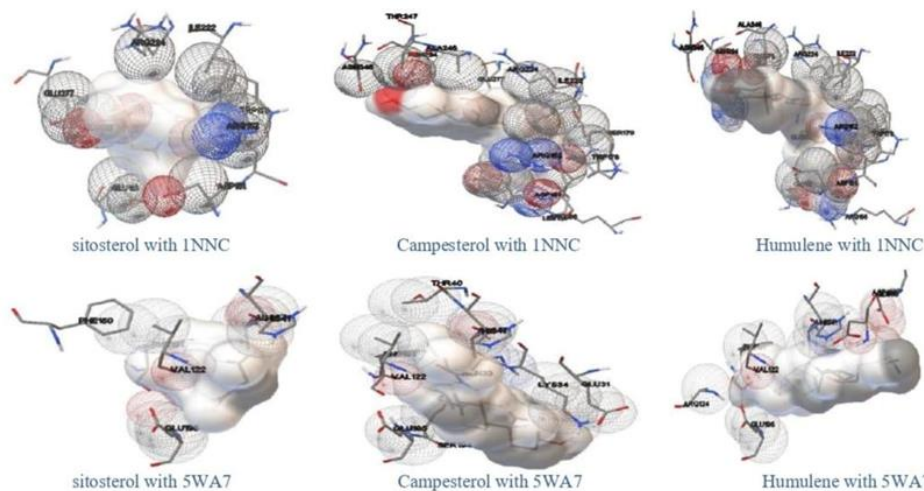


figure 9: Docking visuals of Anti-flu activity

Target protein preparation

The selected proteins from specific organism are downloaded in pdb format. Open it in discovery

biovisualizer and delete the unwanted chains. The unwanted chains and atoms which includes ligand group, repeated chains and non active sites. Then the finalized protein molecule saved in pdb format. In AutoDockToolsTools, Add polar hydrogens then Add Kollman charges and Save as PDBQT (rigid macromolecule)

Molecular Docking Study

a) Grid Generation (AutoGrid)

- Load both protein and ligand.

- Set grid box around active site, set default grid parameters
- Save in .gpf file format.
- Run autogrid4 command in cygwin
- Results in Producing grid map files

b) Docking (AutoDock)

- Load both protein and ligand
- Use Lamarckian Genetic Algorithm (LGA)
- Save .dpf file
- Run autodock4 command in cygwin
- Results in output file: .dlg (docking log file)

Interaction Visualization and result analysis

The resultant .dlg file opened for the docking score. And also open that file in autoDock tool and load protein in order to visualize the interactions between protein and ligand.

Data Interpretation

Docking scores and interaction patterns of Leucasaspera phytochemicals were compared to evaluate their relative binding efficiency. Compounds showing strong binding affinity was considered as potential lead molecules.

RESULTS AND DISCUSSION

For anti-Rheumatoid arthritis

A chronic autoimmune disease where the immune system attacks synovial joints, causing pain, swelling, stiffness, and joint destruction. Usually affects small joints symmetrically.

Target protein for molecular docking,

- Protein 1 – Interleukin-6: Promotes joint inflammation, autoantibody production, and bone resorption.
- Protein 2 – TNF- alpha: Responsible for synovial inflammation, cartilage destruction, and bone erosion.

For anti-Rheumatoid activity, the phytochemicals are docked with both proteins namely Interleukin-6 and TNF- alpha. Among them on docking with protein 1 & 2, the Sitosterol, Campesterol and Baicalein are considered as an most best, where more negative binding energy and lower inhibition constant (Ki) values indicate stronger ligand–protein interactions. Sitosterol showed the highest affinity with binding energies of -7.61 kcal/mol (1ALU) and -9.96 kcal/mol (2AZ5) with ki of 2.64 μ M and 50.11 nm respectively. Campesterol showed -7.42 kcal/mol (1ALU) and -9.58 kcal/mol (2AZ5) with respective ki of 3.64 μ M and 94.41 nM. Baicalein showed -6.66 kcal/mol (1ALU) and -6.69 kcal/mol (2AZ5) with respective ki 13.14 μ M and 12.40 μ M. Overall, the results highlight plant phytosterols, particularly sitosterol and campesterolas promising lead molecules for further pharmacological and experimental validation.

For Anti-asthma

A chronic inflammatory airway disease characterized by reversible bronchoconstriction, airway hyperresponsiveness, and mucus overproduction.

Target protein for molecular docking,

- Protein 1 – B₂-Adrenergic Receptor: Stimulation causes bronchodilation, relieving airway obstruction.
- Protein 2 – Phosphodiesterase (PDE): Inhibition increases cAMP levels leading to bronchial smooth muscle relaxation and reduced inflammation.

For anti-asthma activity, the phytochemicals are docked with both proteins namely Beta 2 -Adrenergic receptor and phosphodiesterase. Among them on docking with protein 1 & 2, the Sitosterol, Campesterol and Baicalein are considered as an most best, where more negative binding energy and lower inhibition constant (Ki) values indicate stronger ligand–protein interactions. Sitosterol showed the highest affinity with binding energies

of -10.43 kcal/mol(2R4R) and -9.17 kcal/mol (1WOJ) with ki of 22.72 nM and 190.89 nm respectively. Campesterol showed -9.43 kcal/mol (2R4R) and -9.18 kcal/mol (1WOJ) with respective ki of 122.97 nM and 186.76 nM. Baicalein showed -7.71 kcal/mol (2R4R) and -7.02 kcal/mol (1WOJ) with respective ki 2.21 μ M and 7.16 μ M. Overall, the results highlight plant phytosterols, particularly sitosterol and campesterolas promising lead molecules for further pharmacological and experimental validation

For Appetite stimulant

Drugs that increase hunger by acting on central appetite-regulating pathways in the hypothalamus. Target proteins for molecular docking,

- Protein 1 – Alpha 2-Adrenergic Receptor: Activation can enhance appetite by reducing sympathetic outflow and modulating hypothalamic feeding signals.
- Protein 2 – GABA-A Receptor: Stimulation increases inhibitory neurotransmission in the CNS, which can promote appetite.

For appetite stimulant activity, the phytochemicals are docked with both proteins namely Alpha 2 -Adrenergic receptor and Dopamine D₂ Receptor. Among them on docking with protein 1 & 2, the Sitosterol, Campesterol and Baicalein are considered as an most best, where more negative binding energy and lower inhibition constant (Ki) values indicate stronger ligand–protein interactions. Sitosterol showed the highest affinity with binding energies of -10.30 kcal/mol (6KUX) and -8.21 kcal/mol (6D6T) with ki of 28.41 nM and 960.31nm respectively. Campesterol showed -9.21 kcal/mol (6KUX) and -8.11 kcal/mol (6D6T) with respective ki of 176.64 nM and 1.13 μ M. Baicalein showed -7.04 kcal/mol (6KUX) and -6.81 kcal/mol (6D6T) with respective ki 6.89 μ M and 10.13 μ M. Overall, the results highlight plant phytosterols, particularly sitosterol and campesterol as promising lead molecules for further pharmacological and experimental validation.

For Anti-inflammatory

Drugs that reduce inflammation by decreasing prostaglandin synthesis.

Target proteins for molecular docking,

- Protein 1 – COX-1: Inhibition reduces prostaglandin formation
- Protein 2 – COX-2: Its inhibition produces the main anti-inflammatory action

For anti-inflammatory activity, the phytochemicals are docked with both proteins namely COX-1 and COX-2. Among them on docking with protein 1 & 2, the Sitosterol, Campesterol and Baicalein are considered as an most best, where more negative binding energy and lower inhibition constant (Ki) values indicate stronger ligand–protein interactions. Sitosterol showed the highest affinity with binding energies of -11.37 kcal/mol (6Y3C) and -11.49 kcal/mol (51KR) with ki of 4.63 nM and 3.80 nm respectively. Campesterol showed -10.91 kcal/mol (6Y3C)

and -11.64 kcal/mol (51KR) with respective k_i of 9.99 nM and 2.92 nM. Baicalein showed -8.24 kcal/mol (6Y3C) and -7.52 kcal/mol (51KR) with respective k_i 911.69 nM and 3.09 μ M. Overall, the results highlight plant phytosterols, particularly sitosterol and campesterol as promising lead molecules for further pharmacological and experimental validation.

For Anti-emetic

Vomiting is triggered by activation of receptors in the chemoreceptor trigger zone (CTZ) and vomiting center. Target proteins for molecular docking,

- Protein 1 – NK₁ (Tachykinin) Receptor: Inhibition prevents emesis.
- Protein 2 – D₂ Receptor: antagonists act as antiemetics.

For anti-emetic activity, the phytochemicals are docked with both proteins namely NK₁ (Tachykinin) and Dopamine D₂ Receptor. Among them on docking with protein 1 & 2, the **Sitosterol, Campesterol and Baicalein** are considered as an most best, where more negative binding energy and lower inhibition constant (K_i) values indicate stronger ligand–protein interactions. Sitosterol showed the highest affinity with binding energies of -9.28 kcal/mol (6E59) and -9.62 kcal/mol (6CM4) with k_i of 156.77 nM and 88.39 nm respectively. Campesterol showed -8.02 kcal/mol (6E59) and -10.68 kcal/mol (6CM4) with respective k_i of 1.33 μ M and 14.97 μ M. Baicalein showed -7.45 kcal/mol (6E59) and -6.47 kcal/mol (6CM4) with respective k_i 3.43 μ M and 18.20 μ M. Overall, the results highlight plant phytosterols, particularly sitosterol and campesterol as promising lead molecules for further pharmacological and experimental validation.

For Anti-malaria

Drugs that kill or inhibit Plasmodium parasites to treat malaria.

Target proteins for molecular docking,

- Protein 1 – Falcipain-2: Its inhibition blocks parasite nutrition and growth.
- Protein 2 – Plasmodium Lactate Dehydrogenase: Inhibition disrupts energy production and leads to parasite death.

For anti-malarial activity, the phytochemicals are docked with both proteins namely falcipain-2 and lactate dehydrogenase. Among them on docking with protein 1 & 2, the Sitosterol, Campesterol and Baicalein are considered as an most best, where more negative binding energy and lower inhibition constant (K_i) values indicate stronger ligand–protein interactions. Sitosterol showed the highest affinity with binding energies of -8.92 kcal/mol (3BPF) and -8.55 kcal/mol (1T24) with k_i of 287.95 nM and 539.00 nm respectively. Campesterol showed -9.87 kcal/mol (3BPF) and -7.26 kcal/mol (1T24) with respective k_i of 57.98 nM and 4.73 μ M. Humulene showed -6.34 kcal/mol (3BPF) and -7.61 kcal/mol (1T24) with respective k_i 22.57 μ M and 2.48 μ M. Overall, the results highlight plant phytosterols,

particularly sitosterol and campesterol as promising lead molecules for further pharmacological and experimental validation.

For Anti-Migraine

A neurovascular disorder causing recurrent throbbing headache due to trigeminal activation and release of vasoactive peptides.

Target proteins for molecular docking,

- Protein 1 – 5-HT_{1B} Receptor: Activation causes cranial vasoconstriction and reduces migraine pain (target of triptans).
- Protein 2 – Calcitonin Gene-Related Peptide: Blocking CGRP (vasodilator released during migraine) helps prevent and treat migraine.

For anti-migraine activity, the phytochemicals are docked with both proteins namely 5HT_{1b} and Calcium gene-related peptides. Among them on docking with protein 1 & 2, the **Sitosterol, Campesterol and Baicalein** are considered as an most best, where more negative binding energy and lower inhibition constant (K_i) values indicate stronger ligand–protein interactions. Sitosterol showed the highest affinity with binding energies of -8.09 kcal/mol (6G79) and -8.21 kcal/mol (7KNU) with k_i of 1.18 μ M and 964.18nm respectively. Campesterol showed -7.42 kcal/mol (6G79) and -7.82 kcal/mol (7KNU) with respective k_i of 3.62 μ M and 1.84 μ M. Baicalein showed -7.37 kcal/mol (6G79) and -6.47 kcal/mol (7KNU) with respective k_i 3.95 μ M and 17.97 μ M. Overall, the results highlight plant phytosterols, particularly sitosterol and campesterol as promising lead molecules for further pharmacological and experimental validation.

For Anti-flu(influenza)

Drugs act by inhibiting key influenza viral enzymes to stop replication and spread.

Target proteins for molecular docking,

- Protein 1 – Neuraminidase: Inhibition blocks release of new viral particles from infected cells, limiting infection spread.
- Protein 2 – PA Endonuclease: Inhibition prevents viral mRNA synthesis, stopping viral replication.

For anti-flu activity, the phytochemicals are docked with both proteins namely Neuraminidase and PA endonuclease. Among them on docking with protein 1 & 2, the sitosterol, campesterol and humulene are considered as an most best, where more negative binding energy and lower inhibition constant (K_i) values indicate stronger ligand–protein interactions. Sitosterol showed the highest affinity with binding energies of -7.79 kcal/mol (1NNC) and -9.96kcal/mol (5WA7) with k_i of 1.94 μ M and 50.11nm respectively. Campesterol showed -7.64 kcal/mol (1NNC) and -9.58 kcal/mol (5WA7) with respective k_i of 2.50 μ M and 94.41nM. Humulene showed -5.90 kcal/mol (1NNC) and -6.85 kcal/mol (5WA7) with respective k_i 47.23 μ M and 9.57 μ M. Overall, the results highlight plant sterols,

particularly sitosterol and campesterols promising lead molecules for further pharmacological and experimental validation.

Here from molecular docking with multiple target protein of various therapeutic activities, among the phytochemical constituents the phytosterols shows greater binding affinity. Then Baicalein and Humulene also shows good affinity towards target protein.

ADMET ANALYSIS

The ADMET analysis of the phytochemical constituents of plant *Leucas aspera* was done with the help of two tools, one is SwissADME predictor by which predicts most absorption, distribution, metabolism and excretion. Another tool is pkCSM by which toxicity also can be predicted. First the smiles of the phytoconstituents are obtained from pubchem software, then pasted it on the smiles space in swissADME page then run for predicted values.

However the swissADME has used to determine absorption, distribution, metabolism and excretion, but shows limitations in toxicity. The pkCSM tool used to determine toxicity studies. As seen already, the smiles of phytochemical constituents are obtained and copied from pubchem and then pasted it in pkCSM online page. Then run it for toxicity predictions.

The ADMET analysis of the phytochemical constituents of plant *Leucas aspera* was done. This analysis

shows that revealed favorable pharmacokinetic properties. Most compounds exhibited high gastrointestinal absorption and complied with Lipinski's rule, indicating good drug-Likeness. The predicted bioavailability ranged from 0.55 to 0.85, suggesting good oral bioavailability. Blood-Brain Barrier permeability was limited to a few compounds, while most were non-BBB permeant.

CONCLUSION

The molecular docking was done with the 15 phytochemical constituents with 16 target protein with respective to their therapeutic activities. From molecular docking studies, the Sitosterol, Campesterol, Baicalein and Humulene are showing greater binding affinity towards target protein. From ADMET analysis, the Baicalein, Thymol, citral shows and greater good properties, the phytochemicals like Humulene, nicotine shows a moderate level of ADMET properties and the phytochemicals like Sitosterol, Campesterol shows a minimal level of properties with some limitations in ADMET properties may the structure modification may improve the properties. The molecular docking studies and ADMET analysis of *Leucas aspera* demonstrated good pharmacokinetic and drug like properties and have a ability to support the therapeutic potential of the plant. This is computational basis for future drug. Discovery effects based on phytochemical of *ophiorrhiza recurvipes* through in vitro and in vivo studies.

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