The Review Of
DIABETIC
STUDIES OPEN ACCESS

In Silico Investigation Of Phytochemicals From Azadirachta Indica, Murraya Koeingii, Aloe Barbadensis Miller And Punica Granatum As Potential Anti-Hyperpigmentation Agents

R.S. Priyengha[1] & Dr. S. Umamaheswari[2]* (Corresponding Author)

[1]Scholar, Department of Biotechnology Manonmaniam Sundaranar University-627012 Tamil Nadu, India.
[2]Professor, Department of Biotechnology,Manonmaniam Sundaranar University-627012Tamil Nadu, India.
Email: umamsu@gmail.com
ORC id: http://orcid.org/0000-0002-5910-5881

Abstract

Hyperpigmentation is a dermatological condition marked by localized overproduction of melanin, often associated with exposure to ultra violet rays, hormonal fluctuations, inflammation, or genetic predisposition. The present study was designed to investigate the anti-hyperpigmentation potential of phytochemicals derived from the methanolic extracts of Azadirachta indica, Aloe barbadensis miller, Punica granatum and Murraya koeingii using GC-MS analysis. A total of 206, 81, 177 and 7 compounds were identified from these plants respectively. Structural information of the identified compounds was retrieved from the PubChem database and pharmacokinetic profiling was conducted using SwissADME. Only compounds satisfying drug-likeness rules and exhibiting favourable skin permeation (log Kp) values were selected for docking studies. A total of 50 compounds passed all the drug-likeness tests of which 14 compounds exhibited good skin permeability. Molecular docking was employed to evaluate the interaction of these 14 phytocompounds using AutoDock with two key protein targets involved in melanogenesis: D-dopachrome tautomerase (PDB ID: 3KAN) and human tyrosinase-related protein 1 (PDB ID: 5M8L). Binding interactions were visualized and analyzed using Biovia Discovery Studio 2024. Two compounds 2,6,10-trimethylundeca-1,3-diene and oxalic acid, cyclobutyl tetradecyl ester exhibited the highest binding affinities against both protein targets, surpassing the docking performance of Thiamidol, a positive drug control. These findings highlight the potential of these naturally occurring compounds as effective inhibitors of melanin synthesis and safer, plant-based alternatives for hyperpigmentation therapy.

1. Introduction

Melanin is an essential pigment and primary determinant of skin, hair and eye colour, is synthesized by specialized organelles called melanosomes through a process known as melanogenesis (Serre et al., 2018). This biochemical process involves a series of multiple melanogenic reactions with tyrosinase (TYR) as the rate-limiting enzyme (Zolghadri et al., 2023). Ultraviolet (UV) exposure stimulates keratinocytes to produce α-melanocyte-stimulating hormone (α-MSH), adrenocorticotropic hormone (ACTH) and endothelin-1. α-MSH binds to melanocortin 1 receptor (MC1R) and increases the level of 3′,5′-cyclic adenosine monophosphate (cAMP). This triggers protein kinase A (PKA) to phosphorylate cAMP response element-binding protein (CREB), which regulates microphthalmia-associated transcription factor (MITF). MITF controls tyrosinase TYR, tyrosinase-related protein 1 (TRP-1) and TRP-2 driving melanogenesis. TYR converts tyrosine to melanin precursors, balancing eumelanin and pheomelanin responsible for black/brown and red/yellow pigmentation respectively. The overactivity or dysregulation of TYR causes hyperpigmentation (Tedasen et al., 2024; D'Mello et al., 2016).

Hyperpigmentation is a common dermatological condition characterized by the excessive production and accumulation of melanin in the skin resulting in skin darkening and uneven skin colour. The condition arises from a complex interplay of factors such as UV radiation exposure, genetic predisposition, injury, hormonal changes and inflammatory responses (Putri et al., 2023). Current hyperpigmentation treatments focus on melanogenesis inhibiting agents like hydroquinone, retinoids, kojic acid, azelaic acid, thiamidol, alpha-

arbutin, niacinamide and vitamin-C (Nautiyal and Wairkar, 2021). However, these synthetic compounds can cause adverse health effects increasing the quest for safer, plant-based alternatives. Many botanical extracts contain flavonoids, polyphenols, terpenoids and alkaloids with melanogenesis-inhibitory properties. Understanding the molecular mechanisms of these compounds may lead to the development of therapeutic strategies with minimal side effects (Veerichetty et al., 2024; Du et al., 2024).

The present study explores the phytochemical composition of Azadirachta indica (Neem), Murraya koeingii (Sweet neem), Aloe b2arbadensis miller (Aloe vera) and Punica granatum (Pomegranate) for their anti-hyperpigmentation activity. Molecular docking was employed to examine how these plant-derived phytochemicals interact with key protein targets involved in melanin synthesis. This research seeks to create safer, plant-derived therapies by utilizing natural inhibitors of melanogenesis to treat hyperpigmentation.

2. Materials and Methods

2.1 Methanolic extraction of Plant Materials

The plant materials (Table 1) were procured from the local market in Thoothukudi, Tamil Nadu and transported to Microbial Biotechnology Laboratory, Manonmaniam Sundaranar University under aseptic conditions. The respective parts of the plant materials were dried separately and powdered coarsely for the extraction process. Methanol was used to extract 50g of dried powder in a Soxhlet apparatus at 65°C. After 8 complete siphon cycles, the extracts were cooled down and filtered through Whatman filter paper to remove impurities. The extracts were dried for 3 days to produce a powder. The crude extract of the plant materials were stored separately in an airtight bottle at room temperature.

Table 1: Plant materials used for the present study, their common name and the part used for the extraction

SI.No	Botanical Name	Common Name (English)	Common Name (Tamil)	Part used
1	Azadirachta indica	Neem	Vembu	Leaf
2	Murraya koeingii	Sweet neem	Karuveppilai	Leaf
3	Aloe barbadensis miller	Aloe vera	Kattralai	Leaf
4	Punica granatum	Pomegranate	Mathulai	Fruit Pulp

2.2 Gas Chromatography-Mass Spectrometry (GC–MS) Analysis

Dried powdered extract of the plant materials were subjected to GC-MS analysis using the Perkin-Elmer Clarus 680 GC system. The apparatus consists of the fused silica column with Elite-5MS (30 m × 250 µm × 0.25 µm). Pure helium (99.9%) was utilized as a carrier gas with 1.0 mL/min constant flow rate and the column was set to operate with the following conditions: the oven was set to operate with an initial temperature of 60°C for 2 min, followed by the constant increase of 10°C per min up to 300°C and the final temperature of 300°C was held for 6 min. The injector temperature was set at 260°C (constant) during the chromatographic run and an injection of 1µL of sample (split ratio 10:1) was used. Perkin-Elmer Clarus 600 EI mass spectrometer was used with a mass-to-charge ratio (m/z) range of 50 to 600 Da. The spectra of the components were compared with the database of spectrum based on retention time and mass spectral patterns of known components stored in the GC-MS NIST (2008) library.

2.3 Ligand Preparation

Structural data of the phytocompounds from the GC-MS analysis were extracted from the PubChem database (https://pubchem.ncbi.nlm.nih.gov). Phytochemical structures from PubChem were downloaded in .sdf format and converted as .pdb format using OpenBabel 2.3 GUI (https://openbabel.org/index.html) which were utilized for molecular docking and pharmacokinetic analysis. Thiamidol (CID: 71543007) was used for the comparative study as a positive control for anti-hyperpigmentation in docking studies.

2.4 Pharmacokinetic Analysis

Drug-likeness and pharmacokinetic properties of each phytocompound were tested with Swiss ADME (http://www.swissadme.ch/). Drug-likeness of compounds was tested with Lipinski's rule of five, Ghose, Veber, Egan and Muegge rules. Other pharmacokinetic properties such as lipophilicity, gastrointestinal absorption, blood brain barrier and skin permeation parameters were evaluated.

2.5 Protein Preparation

The D-dopachrome tautomerase (PDB ID: 3KAN) and human TRP-1 (PDB ID: 5M8L) were downloaded from the protein data bank (https://www.rcsb.org/) in .pdb format. The addition of hydrogen bonds and Kollman charges was performed using the MGL AutoDock Tools 1.5.6.

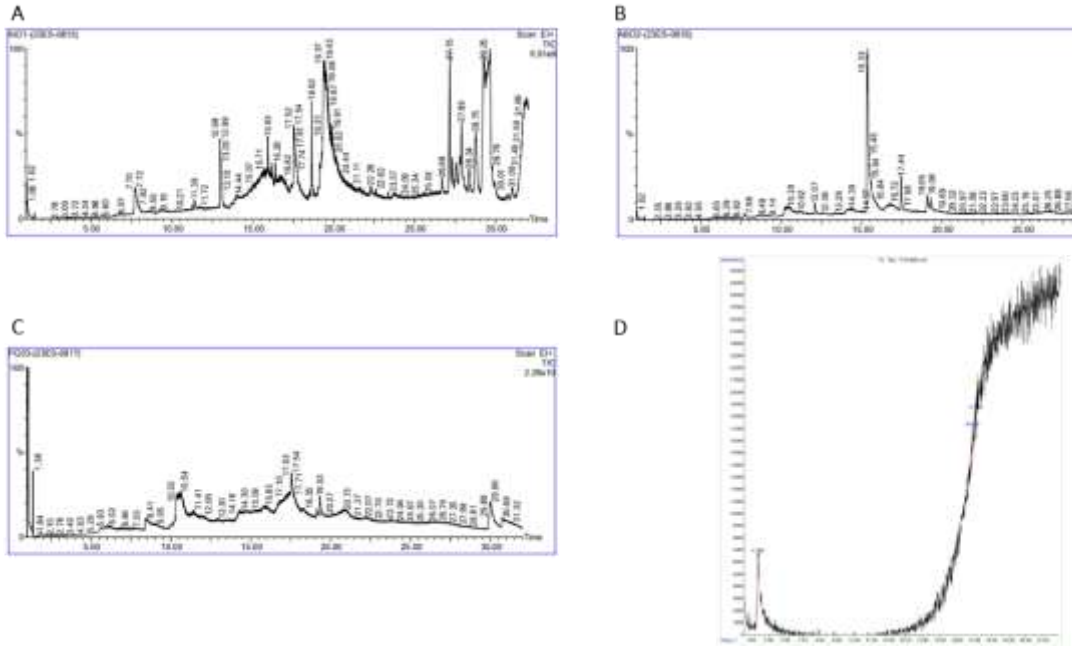
2.6 Molecular Docking

Phytocompounds that have passed Lipinski's rule and have good skin permeability were selected for molecular docking. Computational docking was performed using a specific grid size for the protein with XYZ dimensions (126 X 126 X 126) and a spacing of 0.5 Å for D-dopachrome tautomerase and 1 Å for human TRP-1. Docking was executed using AutoDock 1.5.6 software (https://autodock.scripps.edu/) with the Lamarckian genetic algorithm and default docking parameters. Protein-ligand complexes were visualized in Biovia Discovery Studio 2024 (https://discover.3ds.com/discovery-studio-visualizer-download) to identify the molecular interactions between them.

3. Results and Discussion

Plants are rich sources of bioactive compounds that can counteract the adverse effects of UV exposure to the skin especially plants with large quantities of phenolic compounds that exerts photoprotective properties. Extracts of Pueraria thunbergiana (Kudzu), Piper betle (Betel), Juniperus communis (Juniper), Rhododendron schlippenbachii (Royal azalea), Nelumbo nucifera (Indian lotus), Caesalpinia sappan (Indian redwood), Nymphaea nouchali (Bluewater lily), Zingiber mioga (Japanese ginger),

Figure 1: **GC-MS chromatogram of (A) A. indica, (B) A. barbadensis miller, (C) P. granatum and (D) M. koeingii.**

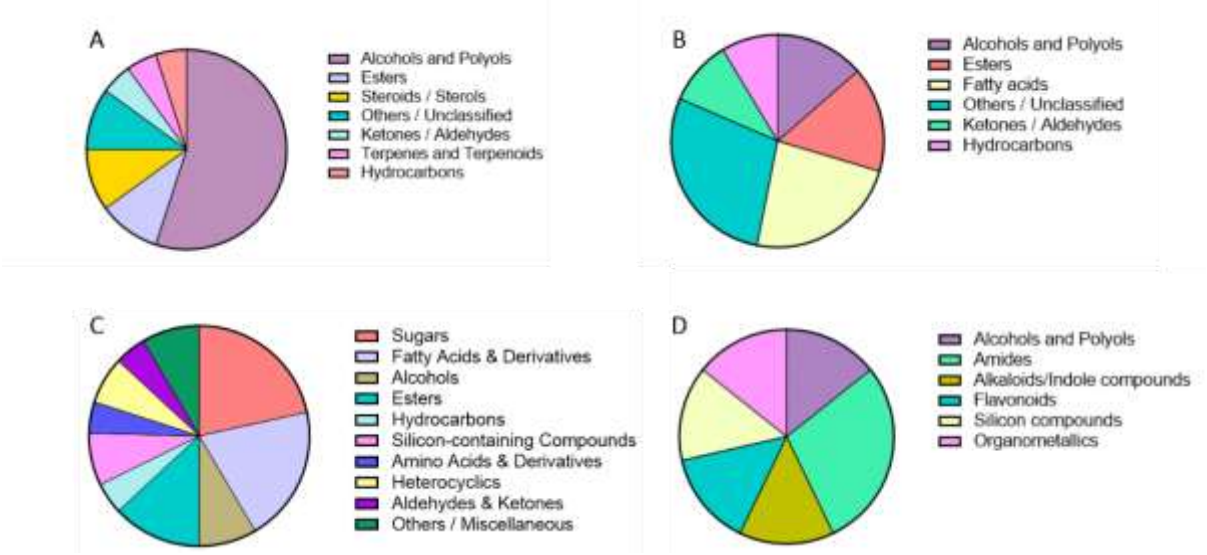


Pyrostegia venusta (Flaming trumpet) and Lespedeza bicolor (Shrubby bushclover) were studied to show effects on melanogenesis and proposed for the treatment of various pigmentation disorders (Gamage et al., 2021; Merecz-Sadowska et al., 2022; Alam et al., 2023). In this present study, methanolic extracts of A. indica, A. barbadensis miller, P. granatum and M. koeingii were used for the GC-MS analysis (Figure **1**).

Around 206 compounds were identified in A. indica which is higher than the compounds derived from its ethanolic extract (Loganathan et al., 2021; Bolade et al., 2018). Among the identified compounds majority

of them belong to alcohols and polyols followed by esters. A total of 81 compounds were identified in the methanolic extracts of A. barbadensis miller which is found to be higher than the compounds identified in its ethanolic extract (Alghamdi et al., 2023). Based upon the identified compounds, most of them are fatty acids and unclassified compounds. Meanwhile, P. granatum has shown higher number of identified compounds (177 compounds) with larger amounts of sugars followed by fatty acids than their ethanolic counterpart (Attia 2019). In the extracts of M. koeingii, 7 compounds were identified and shown to be lesser than its ethanolic extract (Azhagu Madhavan et al., 2021; Wirjosentono et al., 2019). The composition and distribution of phytocompounds identified through GC-MS are shown in Figure 2.

Figure 2: Distribution of GC-MS identified compounds of (A) A. indica, (B) A. barbadensis miller, (C) P. granatum and (D) M. koeingii based upon their nature.



After the complete identification of the phytocompounds using GC-MS, their 3D structural data were retrieved from PubChem database. PubChem database is a public repository for information on the physical, chemical, structural and biological properties of chemical compounds (Kim et al., 2016). Their structural data was used to study the pharmacokinetic and drug-likeness properties in SwissADME. SwissADME predicts absorption, distribution, metabolism and excretion (ADME) parameters aiding in the identification of potential drug candidates (Daina et al., 2017). Based on the results (Supplementary data 1), 24 compounds in A. indica, 14 compounds in A. barbadensis miller, 9 compounds in P. granatum and 3 compounds in M. koeingii have passed all the drug-likeness rules. Compounds that have passed all the drug-likeness rules with good skin permeation (log Kp) were selected for the molecular docking analysis.

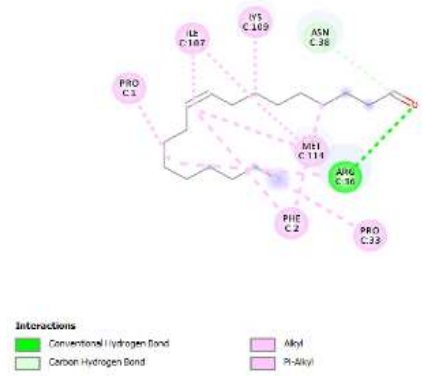
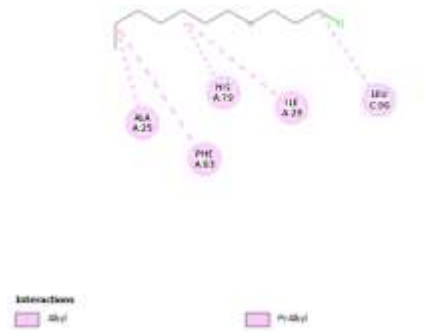
Out of the 50 potential drug candidates, 14 phytocompounds have good skin permeability and thus can penetrate the skin layers to exert effects on melanogenesis. Based on a wide literature search, two protein targets (D-dopachrome tautomerase; human TRP-1) and one positive control drug (Thiamidol) were selected. Both D-dopachrome tautomerase and TRP-1 plays a pivotal role in the biosynthesis of melanin. In hyperpigmentation disorders, TRP-1 activity accelerates excessive melanin production, making it a promising drug target for skin-lightening agents and therapeutic interventions against pigmentation-related conditions. (Milac et al., 2017; Lai et al., 2018). Thiamidol is an active chemical compound known as Isobutylamido-Thiazolyl-Resorcinol used as a depigmenting agent in skincare to reduce hyperpigmentation. It works by selectively inhibiting TYR, making it effective in treating post-inflammatory hyperpigmentation, melasma, UVB-induced hyperpigmentation and discoloration (Desai et al., 2025; Schuster and Sammain 2024).

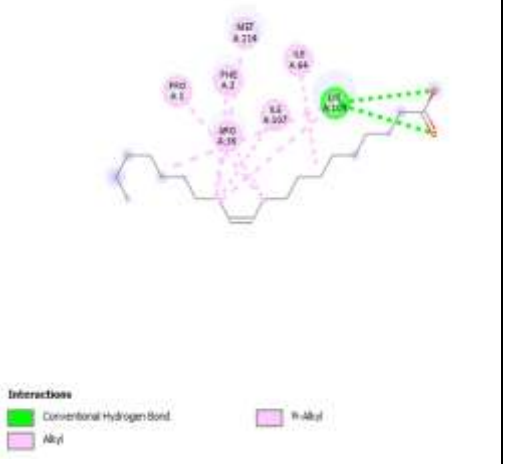
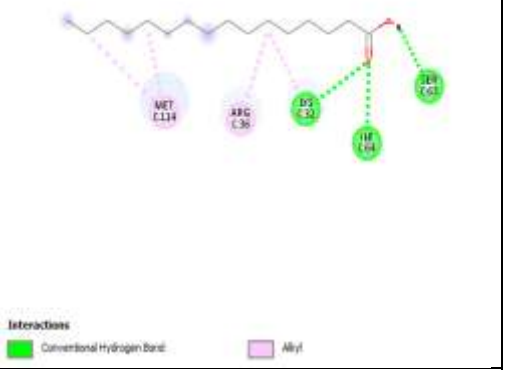
AutoDock is a free, user-friendly widely used computational tool for molecular docking (MD) to study the interaction between a receptor and a ligand. It aids in the identification of binding conformations, binding modes, binding sites and the calculation of binding affinities of a receptor-ligand complex. It has a major application in structure-based drug designing, virtual screening and drug discovery (Morris et al., 2008).

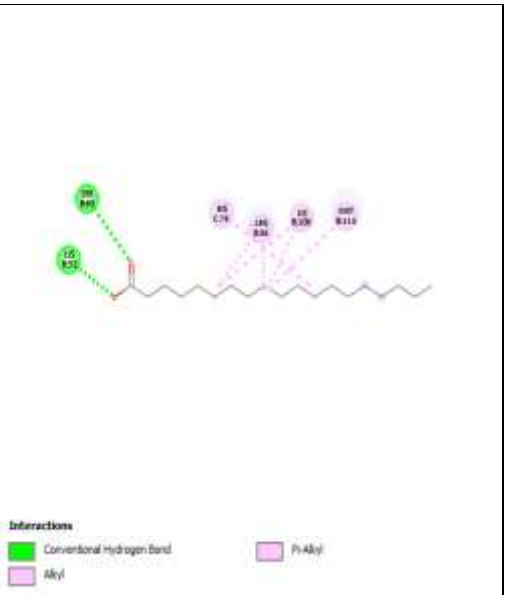
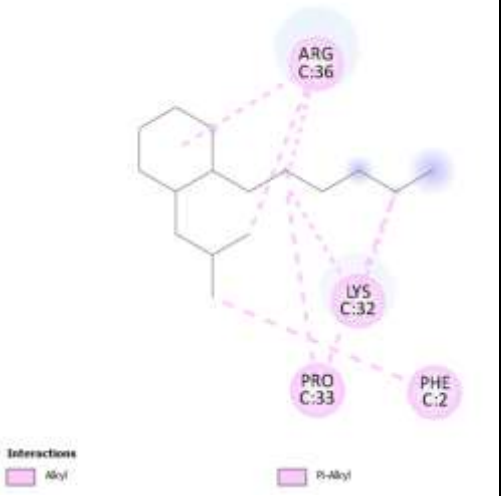
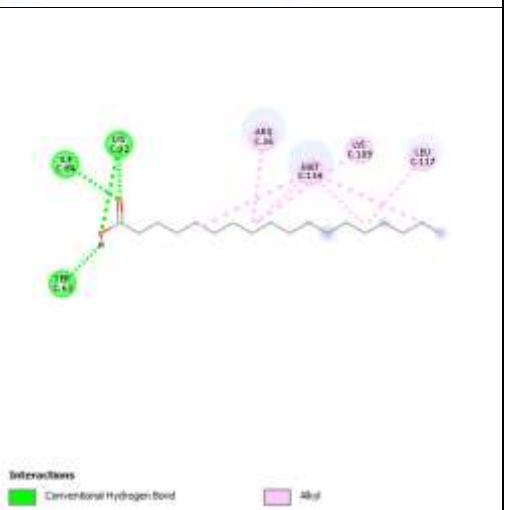
Docking was performed for the 14 phytocompounds along with 1 positive control drug and visualised in Biovia Discovery Studio 2024.

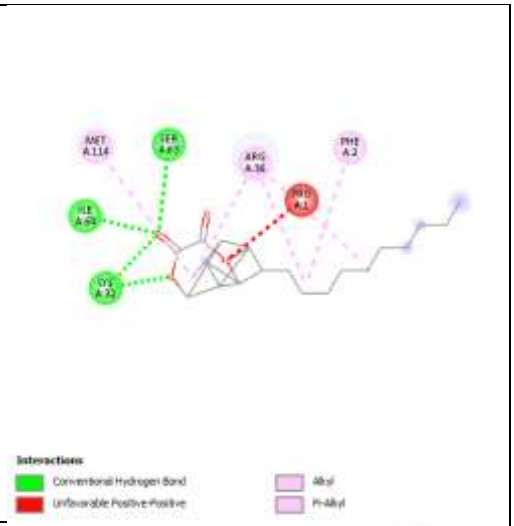
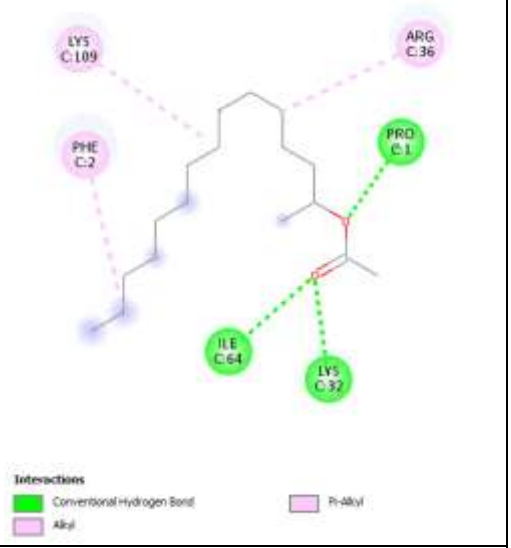
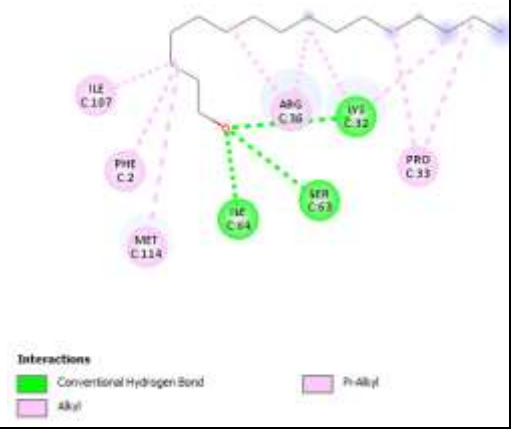
Based on the MD results (Table 2), 2,6,10-trimethylundeca-1,3-diene, Octadecanoic acid, Oxalic acid cyclobutyl tetradecyl ester, N-Hexadecanoic acid and Nonadecanoic acid indicated higher binding energy with D-dopachrome tautomerase than thiamidol (-6.27). When compared with thiamidol, based on hydrogen bond counts, Octadecanoic acid and Oxalic acid cyclobutyl tetradecyl ester have exhibited the same stable interactions with the protein target. Even though 2,6,10-trimethylundeca-1,3-diene (-8.26) has higher binding affinity, it does not have any hydrogen bonds resulting in poor stability. Notably, 2 phytocompounds: 6-ethyl-3-di(tert-butyl)silyloxyoctane and 3-Dimethylsilyloxytridecane has shown the absence of any interaction with the target protein.

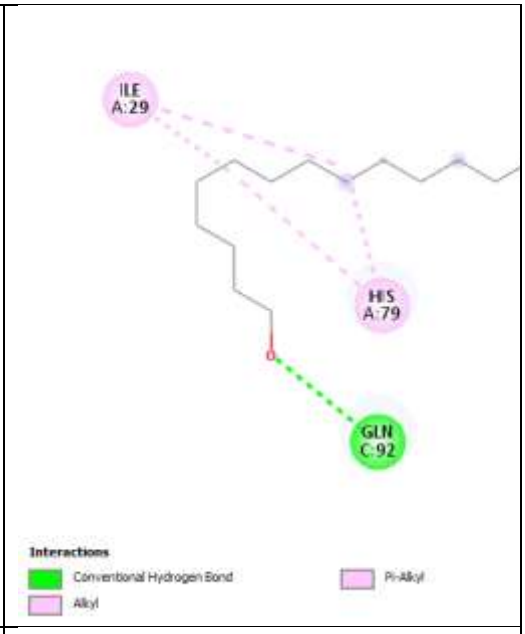
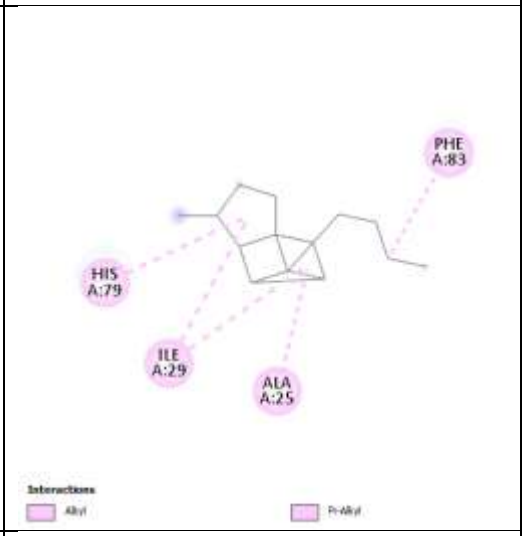
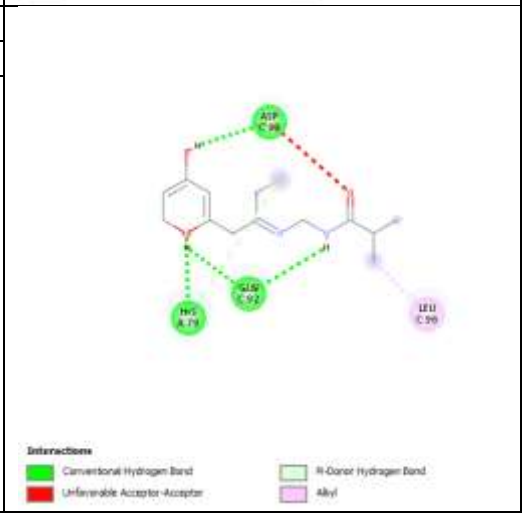
Table 2: Molecular docking of phytocompounds and Thiamidol with D-dopachrome tautomerase.

SI.No	Ligand	Binding Energy (kcal/mol)	Ligand – Protein Interaction (Hydrogen Bond)	Distances	2D Image of the interaction
1	9-Octadecenal, (Z)-	-5.10	ARG C:36	2.86	
			ASN C:38	2.98	
2	1-Chloroundecane	-4.61	-	-	

3	3-Dimethylsilyloxytridecane	+1.97	-	-	-
4	Cis-11-eicosenoic acid	-7.01	LYS A:109	2.6 2.73	
5	N-Hexadecanoic acid	-7.09	LYS C:32	2.83	
			ILE C:64	2.8	
			SER C:63	1.96	
6	Nonadecanoic acid	-7.08	SER B:63	3.30	

			LYS B:32	2.63	
7	Cis-1-methyl-3-N-nonylcyclohexane	-6.95	-	-	
8	Octadecanoic acid	-7.8	LYS C:32	3.25 2.62	
			ILE C:64	3.24	
			SER C:63	2.11	
9		-7.65	SER A:63	3.22	
			ILE A:64	2.66	

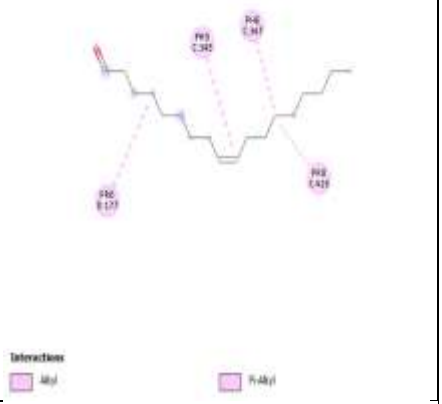
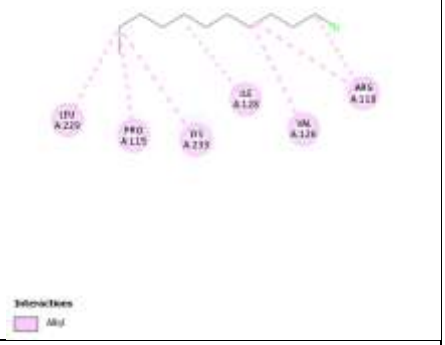
	Oxalic acid, cyclobutyl tetradecyl ester		LYS A:32	2.88 3.04	
10	2-Pentadecanol acetate	-5.41	PRO C:1 ILE C:64 LYS C:32	2.96 2.8 2.87	
11	6-ethyl-3-di(tert-butyl)silyloxyoctane	+1.97	-	-	-
12	Oxirane, tetradecyl-	-5.31	ILE C:64 SER C:63 LYS C:32	2.72 3.11 3.06	

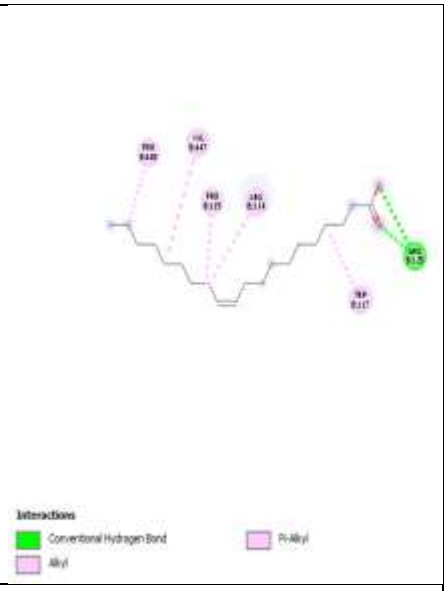
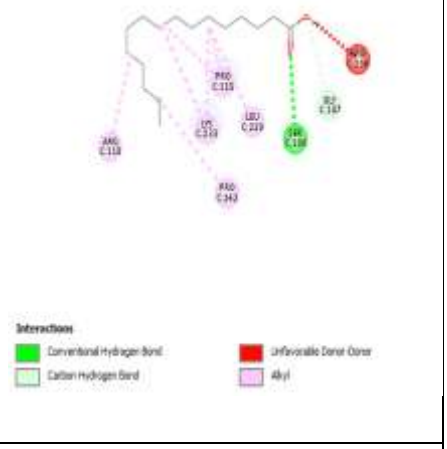
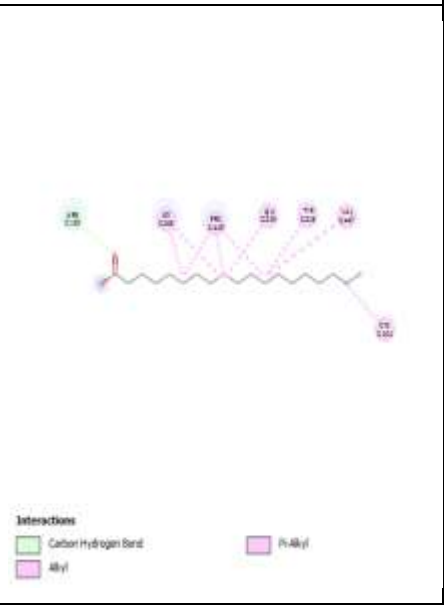
13	Oxirane, tridecyl-	-5.13	GLN C:92	2.88	
14	2,6,10-trimethylundeca-1,3-diene	-8.26	-	-	
15	Thiamidol	-6.72	ASP C:93 HIS A:79 GLN C:92	2.10 2.94 1.90 2.44	

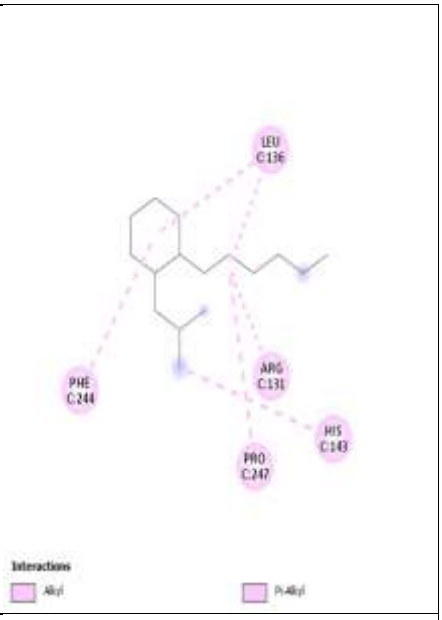
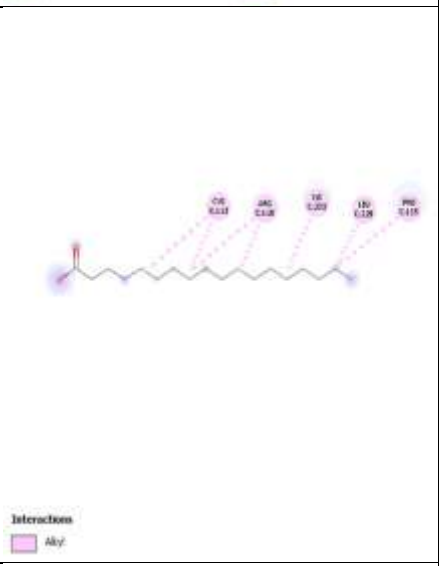
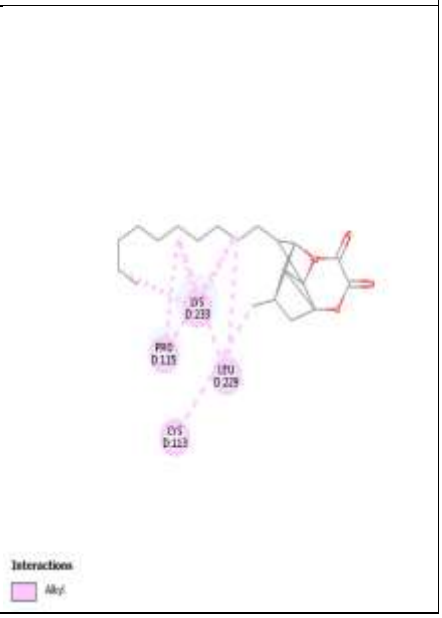
Molecular docking results (Table 3) of phytocompounds and Thiamidol with human TRP-1 show 2,6,10-trimethylundeca-1,3-diene, Oxalic acid cyclobutyl tetradecyl ester and Cis-1-methyl-3-N-nonylcyclohexane indicated higher binding energy than positive drug control (-4.57). Whereas when compared with the number of hydrogen bonds, Thiamidol has four and these above-mentioned compounds

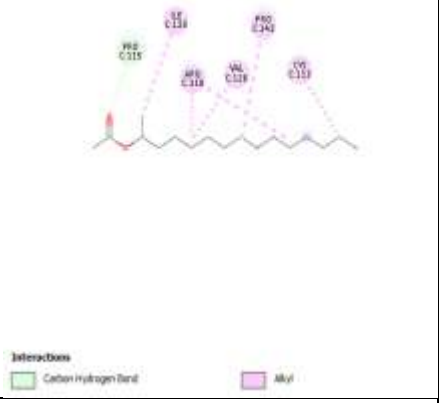
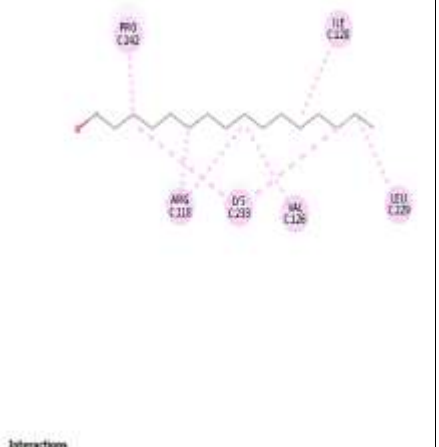
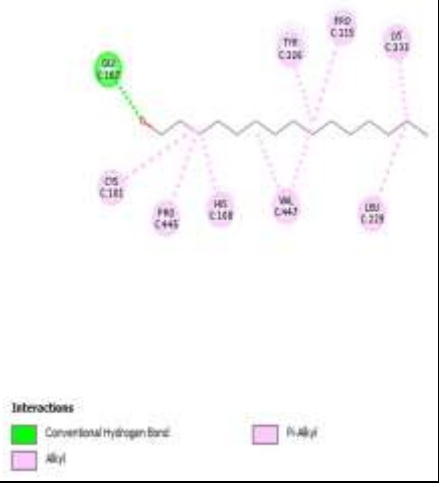
had none. Also 6-ethyl-3-di(tert-butyl)silyloxyoctane and 3-Dimethylsilyloxytridecane have shown the absence of any interaction with the target protein.

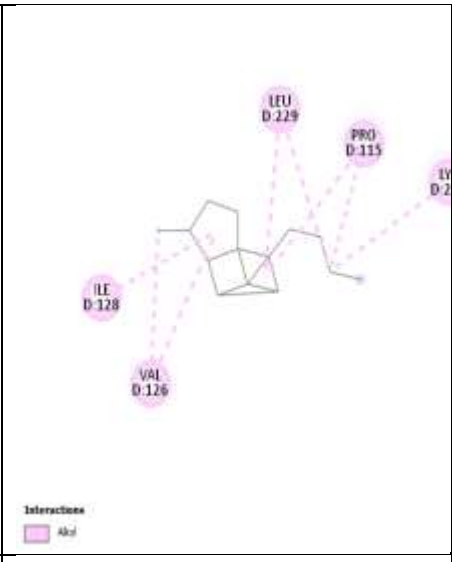
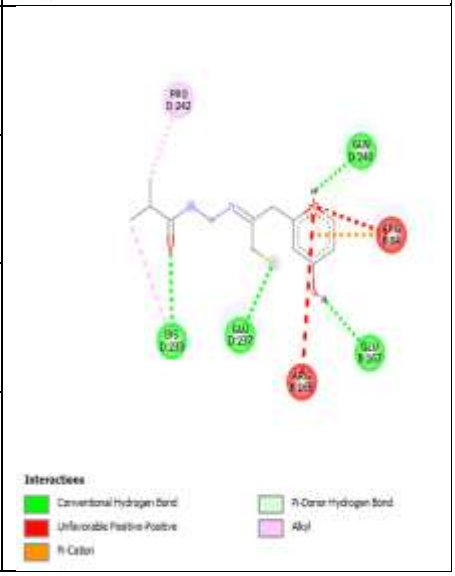
Table 3: Molecular docking of phytocompounds and Thiamidol with human tyrosinase related protein 1.

SI.No	Ligand	Binding Energy (kcal/mol)	Ligand – Protein Interaction (Hydrogen Bond)	Distances	2D Image of the interaction
1	9-Octadecenal, (Z)-	-2.40	-	-	
2	1-Chloroundecane	-3.9	-	-	
3	3-Dimethylsilyloxytridecane	+1.95	-	-	-

4	Cis-11-eicosenoic acid	-2.2	ARG B:125	3.16 3.04	
5	N-Hexadecanoic acid	-2.83	SER C:106	2.95	
6	Nonadecanoic acid	-2.48	ARG C:118	3.26	

7	Cis-1-methyl-3-N-nonylcyclohexane	-5.06	-	-	
8	Octadecanoic acid	-2.01	-	-	
9	Oxalic acid, cyclobutyl tetradecyl ester	-5.89	-	-	

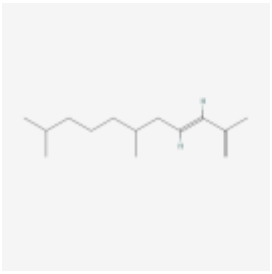
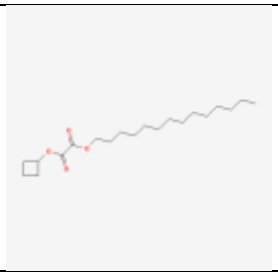
10	2-Pentadecanol acetate	-3.79	PRO C:115	3.49	
11	6-ethyl-3-di(tert-butyl)silyloxyoctane	+1.96	-	-	-
12	Oxirane, tetradecyl-	-3.58	-	-	
13	Oxirane, tridecyl-	-3.22	GLY C:107	2.73	

14	2,6,10-trimethylundeca-1,3-diene	-6.83	-	-	
15	Thiamidol	-4.57	LYS D:233	3.17	
			GLU D:23	3.4	
			GLU B:16	2.38	
			GLN D:24	2.44	

To conclude based on the results, with both the protein targets, 2,6,10-trimethylundeca-1,3-diene and Oxalic acid, cyclobutyl tetradecyl ester showed higher binding energy. Elaborative information on these compounds, their structure, binding energy and other properties have been tabulated in Table 4. Furthermore, the results highlight the presence of hydrophobic interactions and underscore the diversity of bonds shared between the ligand-protein complex indicating potential activity against melanogenesis.

Table 4: **Summary of molecular docking results of 2,6,10-trimethylundeca-1,3-diene and Oxalic acid, cyclobutyl tetradecyl ester.**

Name of the Phytocompound	**Chemical Structure**	**Binding Energy**		**No. of Bonds**		**Log Kp value for Skin Permeation**	**No. of Drug-likeness rules passed**
		3KAN	**5M8L**	**3KAN**	**5M8L**		

2,6,10-trimethylundeca-1,3-diene		-8.26	-6.83	5	8	-2.98 cm/s	4
Oxalic acid, cyclobutyl tetradecyl ester		-7.65	-5.89	9	8	-2.71 cm/s	3

4. Conclusion

Phytocompounds isolated from the methanolic extracts of A. indica, A. barbadensis miller, P. granatum and M. koeingii have various bioactive properties. Only a handful have passed the rigorous filtering in the present study. Both 2,6,10-trimethylundeca-1,3-diene and Oxalic acid, cyclobutyl tetradecyl ester demonstrate a notable affinity for the target proteins associated with melanogenesis. Notably, they exert higher binding energy than the conventional drug Thiamidol. This study highlights the phytocompounds as an effective anti-hyperpigmentation agent, primarily by inhibiting enzymatic activities important for melanin synthesis and lays the groundwork for future in vitro and in vivo validation, paving the way towards the development of novel cosmeceuticals derived from medicinal plants.

5. References

1. Serre C, Busuttil V, Botto JM. Intrinsic and extrinsic regulation of human skin melanogenesis and pigmentation. International Journal of Cosmetic Science. 2018 Aug;40(4):328-47.
2. Zolghadri S, Beygi M, Mohammad TF, Alijanianzadeh M, Pillaiyar T, Garcia-Molina P, Garcia-Canovas F, Munoz-Munoz J, Saboury AA. Targeting tyrosinase in hyperpigmentation: Current status, limitations and future promises. Biochemical pharmacology. 2023 Jun 1;212:115574.
3. Tedasen A, Chiabchalard A, Tencomnao T, Yamasaki K, Majima HJ, Phongphithakchai A, Chatatikun M. Anti-melanogenic activity of ethanolic extract from Garcinia atroviridis fruits using in vitro experiments, network pharmacology, molecular docking, and molecular dynamics simulation. Antioxidants. 2024 Jun 12;13(6):713
4. D'Mello SA, Finlay GJ, Baguley BC, Askarian-Amiri ME. Signaling pathways in melanogenesis. International journal of molecular sciences. 2016 Jul 15;17(7):1144.
5. Putri LA, Anjani NL, Laksmiani NP, Susanti NM. In silico molecular docking of luteolin as a potential antihyperpigmentation agent. Pharmacy Reports. 2023;3(1):61.
6. Nautiyal A, Wairkar S. Management of hyperpigmentation: Current treatments and emerging therapies. Pigment cell & melanoma research. 2021 Nov;34(6):1000-14.
7. Veerichetty V, Sivaraman L, Jeyasaravanan S, Dharmalingam T, Almoallim HS, Aljawdah HM. Evaluation of Pomegranate Seed Extract as a Tyrosinase Inhibitor for Hyperpigmentation Treatment. Indian Journal of Pharmaceutical Education and Research. 2024 Jun 21;58(3):965-75.
8. Du R, Ye L, Chen X, Meng Y, Zhou L, Chen Q, Zheng G, Hu J, Shi Z. Screening of Key Components for Melanogenesis Inhibition of Polygonum cuspidatum Extract Based on the Spectrum–Effect Relationship and Molecular Docking. Molecules. 2024 Feb 15;29(4):857.
9. Gamage DG, Dharmadasa RM, Abeysinghe DC, Wijesekara RG, Prathapasinghe GA, Someya T. Ethnopharmacological survey on medicinal plants used for cosmetic treatments in traditional and ayurveda systems of medicine in Sri Lanka. Evidence-Based Complementary and Alternative Medicine. 2021;2021(1):5599654.

10. Merecz-Sadowska A, Sitarek P, Stelmach J, Zajdel K, Kucharska E, Zajdel R. Plants as modulators of melanogenesis: Role of extracts, pure compounds and patented compositions in therapy of pigmentation disorders. International Journal of Molecular Sciences. 2022 Jan;23(23):14787.
11. Alam MB, Park NH, Song BR, Lee SH. Antioxidant potential-rich betel leaves (Piper betle L.) exert depigmenting action by triggering autophagy and downregulating MITF/tyrosinase in vitro and in vivo. Antioxidants. 2023 Feb 3;12(2):374.
12. Loganathan T, Barathinivas A, Soorya C, Balamurugan S, Nagajothi TG, Jayakumararaj R. GCMS profile of bioactive secondary metabolites with therapeutic potential in the ethanolic leaf extracts of Azadirachta indica: A sacred traditional medicinal plant of india. Journal of Drug Delivery and Therapeutics. 2021 Jul 2;11(4-S):119-26.
13. Bolade OP, Akinsiku AA, Adeyemi AO, Williams AB, Benson NU. Dataset on phytochemical screening, FTIR and GC–MS characterisation of Azadirachta indica and Cymbopogon citratus as reducing and stabilising agents for nanoparticles synthesis. Data in brief. 2018 Oct 1;20:917-26.
14. Alghamdi A, Alshehri W, Sajer B, Ashkan M, Ashy R, Gashgari R, Hakmi H. Biological Activities and GC-MS Analysis of Aloe vera and Opuntia ficus-indica Extracts. Journal of chemistry. 2023;2023(1):6504505.
15. Attia EA. Antimicrobial Activity and Bio-active compounds analysis in Ethanolic plant extracts of Punica Grantanum (Pomegranate) using GC-MS. Egypt J. Exp. Boil. 2019 Jul 1;15:325.
16. Azhagu Madhavan S, Sa V, Ra S, Sb R. Phytochemical screening and GC–MS analysis of bioactive compounds present in ethanolic leaf extract Murraya koenigii. Bull Env Pharmacol Life Sci. 2021 Mar 4;10:158-64.
17. Wirjosentono B, Marpaung L. Phytochemical screening and chemical analysis of ethanol extract of Kari leaves (murayya koeginii) using GC-MS method. InJournal of Physics: Conference Series 2019 Sep 1 (Vol. 1232, No. 1, p. 012012). IOP Publishing.
18. Kim S, Thiessen PA, Bolton EE, Chen J, Fu G, Gindulyte A, Han L, He J, He S, Shoemaker BA, Wang J. PubChem substance and compound databases. Nucleic acids research. 2016 Jan 4;44(D1):D1202-13.
19. Daina A, Michielin O, Zoete V. SwissADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. Scientific reports. 2017 Mar 3;7(1):42717.
20. Milac AL, Negroiu G. The multiple roles of tyrosinase-related protein-2/L-Dopachrome tautomerase in melanoma: biomarker, therapeutic target, and molecular driver in tumor progression. InHuman Skin Cancers-Pathways, Mechanisms, Targets and Treatments 2017 Dec 20. IntechOpen.
21. Lai X, Wichers HJ, Soler-Lopez M, Dijkstra BW. Structure and function of human tyrosinase and tyrosinase-related proteins. Chemistry–A European Journal. 2018 Jan 2;24(1):47-55.
22. Desai S, Alexis A, Baldwin H, Callender V, Elbuluk N, Farris P, Grimes P, Taylor S, Frey C, Romanowitz D. Thiamidol: A Breakthrough Innovation in Treatment of Hyperpigmentation. SKIN The Journal of Cutaneous Medicine. 2025 Mar 17;9(2):s548-.
23. Schuster B, Sammain A. Hyperpigmentation in skin of colour: Therapeutical benefits of isobutylamido-thiazolyl-resorcinol (Thiamidol®), an effective tyrosinase inhibitor, in phototypes IV–VI. JEADV Clinical Practice. 2024 Jul;3(3):801-6.
24. Morris GM, Huey R, Olson AJ. Using autodock for ligand-receptor docking. Current protocols in bioinformatics. 2008 Dec;24(1):8-14.