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Hypoglycemic, anti-inflammatory, and anti-aging potential of *Canthium coromandelicum* (Burm.f.) Alston leaf extracts: *In vitro* and *in silico* ADMET studies

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ABSTRACT

The present study focused on the hypoglycemic, anti-inflammatory and anti-aging effects of *Canthium coromandelicum* (Burm.f.) Alston leaf extract by *in vitro* and *in silico* analyses. *In vitro* antidiabetic activities were studied by α-amylase and α-glucosidase enzyme inhibition assays. The anti-inflammatory activity was analysed by bovine serum albumin denaturation assay and anti-aging activity by the inhibition of tyrosinase enzyme. The molecular docking study was undertaken to examine the binding mode and interactions of reported compounds of the hydroalcoholic extract of *C. coromandelicum* leaf with 7TAA, 3WY1, 2Y9X, and 4COX proteins. The plant exhibited promising antidiabetic effects with an inhibition of 86.09% against α-amylase and 83.53% against α-glucosidase enzymes. The extract had a notable anti-inflammatory activity with an inhibition range of 50.23 to 73.17%. The anti-aging activity showed 50.31 to 76.78% of inhibition in anti-tyrosinase assay with a potential effect in hyperpigmentation. In molecular docking analysis, the compounds like linoleic acid, phytol, methyl linoleate, palmitic acid and ethyl octa-decanoate had potent inhibitory activities against 7TAA, 3WY1, 2Y9X and 4COX proteins (6.90 to -4.42 kcal/mol) especially linoleic acid was found to be highly effective (-6.11 kcal/mol) antidiabetic effect with notable binding scores. The obtained results suggest that phytoconstituents identified from the plant have the potential in controlling diabetes by inhibiting α-amylase and α-glucosidase enzymes with promising anti-inflammatory and anti-aging properties. However, further research is required to ascertain its potential effect in experimental animals and isolation of bioactive compounds.

1. Introduction

Diabetes mellitus is one of the metabolic disorders categorized by the resistance to insulin action, *i.e.*, insufficient secretion of insulin. The α-amylase and α-glucosidase enzymes are responsible for slowed release of glucose from starch and oligosaccharides. This causes a delay in absorption of glucose and thus leading to reduced postprandial blood glucose levels (Cisneros-Yupanqui et al., 2023). Plants produce a large number of compounds such as α-amylase and α-glucosidase inhibitors which are able to regulate the activities of these enzymes (Dwivedi et al., 2021). Inflammation is a pathophysiological process regulated by infinite signaling molecules produced by leukocytes and macrophages that can be treated with traditional medicines with significant success and relief without any side effects (Nigussie et al., 2021). Cyclooxygenase (COX) is an enzyme that catalyses the production of prostaglandin hormone which has significant role in inflammation (Khan et al., 2022). Tyrosinase is one of the polyphenolic oxidase compounds involved in forming specific pigments (melanin), browning of several types of fruits and vegetables by enzymatic reactions (Ali et al., 2022). Tyrosinase inhibitors can be used in treating skin hyperpigmentation and also assist with anti-aging, hence research efforts aimed at identifying safer and more potent stable tyrosinase inhibitors (Seo et al., 2021).

Several factors are involved in pathophysiology of diabetes including inflammation and related complications. Many pro-inflammatory cytokines play a critical role in the development and progression of diabetes and its complications through low-grade inflammation (Henein et al., 2022). Aging is another significant factor in diabetes and it is associated with raised levels of pro-inflammatory

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molecules, including interleukin (IL) 1, IL-6, IL-8, IL-13, IL-18, interferons α and β, transforming growth factor β and tumor necrosis factor-alpha (Mishra et al., 2022). In recent years, researchers have established the glucose lowering effect of phytocompounds like lupeol and isoorientin with molecular docking simulation which simplifies *in vivo* and *in vitro* studies in drug modeling (Batool et al., 2019; Seo et al., 2021). *In silico* studies have emerged as one of the pertinent methods to perceive the drug responses using diverse computational tools (Seo et al., 2021). The main purpose of molecular docking is to predict the affinity of drugs or ligands to bind with target proteins. The structure-based drug design has been developed as a low-cost strategy to improve the success rate at any stage of the drug discovery process while using the protein-ligand docking (Khanal et al., 2022; Sapkal et al., 2023).

The hydroalcoholic extract of *Canthium coromandelicum* (Burm.f.) Alston (family Rubiaceae) leaves were selected in this study based on its medicinal properties. Various parts of the plant species are used to treat cough, diarrhea, diabetes, fever, indigestion, inflammations, skin diseases and snake bites (Mahishi et al., 2005; Amalraj et al., 2021). In Ayurvedic medicine, it has been used to treat a variety of ailments including fever, skin diseases, digestive disorders and respiratory problems. It has also been used to treat urinary tract infections, skin infections, gastrointestinal infections, malaria and dengue fever, liver disorders such as jaundice and hepatitis (Rana et al., 2023). The present study was undertaken to examine the *in vitro* hypoglycemic, anti-inflammatory, and anti-aging effects of the hydroalcoholic extract of *C. coromandelicum* leaf followed by *in silico* studies. The major compounds (ligands) used for docking study (Table 1 and Fig. 1) were retrieved from the previous GC–MS analysis report (Amalraj et al., 2021). The 3D crystal structure of selected proteins, 7TAA, 3WY1, 4COX, and TY9X were used for the molecular docking studies.

2. Materials and methods

2.1. Chemicals and reagents

Sodium phosphate monobasic, Sodium phosphate dibasic, sodium chloride, dinitrosalicylic acid, p-nitrophenyl-α-dglucopyranoside, bovine serum albumin, tyrosinase, L-DOPA, diclofenac, kojic acid, acarbose, α-amylase and α-glucosidase enzymes were purchased from Sisco Research Laboratories Pvt. Ltd, Sigma-Aldrich and Merck Life Science Private Limited (Mumbai, India).

2.2. Collection and extraction of plant material

The leaves of *C. coromandelicum* were collected from the wild habitats of the species in Thanjavur district, India in September 2020. After the plant specimens were positively identified as *C. coromandelicum*, a voucher specimen was prepared and preserved in the Herbarium, Department of Botany, A.V.V.M. Sri Pushpam College (SPCH-902). Shade dried leaves were ground into powder using an electric blender and kept in airtight containers. For extraction, 50 g of powdered leaf were soaked in 500 mL of hydroalcohol (ethanol 70%:aqueous 30%) in conical flask for 24 h at room temperature. The crude extract of *C. coromandelicum* leaf was filtered through Whatmann No.1 filter paper, and then stored in a deep freezer at 4 °C for further studies.

2.3. Hypoglycemic effect C. coromandelicum leaf extract

Hypoglycemic effect of the *C. coromandelicum* leaf extract was carried out by α-amylase and α-glucosidase enzyme inhibition assays as detailed in the previous report (Amalraj et al., 2021) using acarbose as positive control and obtained results were calculated as follows:

$$\% \text{ of inhibition} = ((\text{Control OD} - \text{Sample OD})/\text{Control OD}) \times 100$$

2.4. Anti-inflammatory activity of C. coromandelicum leaf extract

Anti-inflammatory activity of *C. coromandelicum* leaf extract was evaluated by bovine serum albumin denaturation assay (Raj et al., 2023). To one mL of extract, 200 μL bovine albumin (1%) and 3.8 mL phosphate buffer saline were added. The mixture was incubated for 15 min at 37 °C. After heating for 5 min, reaction mixture was cooled in room temperature, then the absorbance was recorded at 660 nm with phosphate buffer saline as control and diclofenac as standard. The inhibition level of protein denaturation was estimated using this formula:

$$\% \text{ of inhibition of sample} = \frac{\text{Control OD} - \text{Sample OD}}{\text{Control OD}} \times 100$$

Table 1
Physicochemical properties of phytocompounds of the hydroalcoholic leaf extract of *C. coromandelicum* accordance with the rules of drug-likeness.

Ligands/Compound Names	MF	MW	Log P	NA	HD	Log	LV	RB
Palmitic acid	$C_{16}H_{32}O_2$	256.42	6.14	2	4	−3.04	1	14
Ethyl octadecanoate	$C_{20}H_{40}O_2$	312.53	7.63	2	1	−1.84	1	18
n-Dodecane	$C_{12}H_{26}$	170.33	7.78	0	0	−3.01	1	9
Heptacosane	$C_{27}H_{56}$	380.73	16.98	0	0	1.49	1	24
n-Tridecane	$C_{13}H_{28}$	184.36	8.39	0	0	−2.7	1	10
n-Docosane	$C_{22}H_{46}$	310.6	13.91	0	0	−0.01	0	19
n-Pentacosane	$C_{25}H_{52}$	352.68	15.75	0	0	0.89	1	22
Hexacosane	$C_{26}H_{54}$	366.71	16.36	0	0	1.19	1	23
Octacosane	$C_{28}H_{58}$	394.76	17.59	0	0	1.79	1	25
n-Tricosane	$C_{23}H_{48}$	324.63	14.52	0	0	0.29	1	20
n-Tetracosane	$C_{24}H_{50}$	338.65	15.14	0	0	0.59	1	21
n-Undecane	$C_{11}H_{24}$	156.31	7.17	0	0	−3.31	0	8
n-Decane	$C_{10}H_{22}$	142.28	6.65	0	0	−3.61	0	7
Ethyl tetradecanoate	$C_{16}H_{32}O_2$	256.42	5.86	1	2	−2.77	1	14
Phytol	$C_{20}H_{40}O$	296.53	6.82	1	2	−2.29	1	13
Linoleic acid	$C_{18}H_{32}O_2$	280.45	5.48	2	4	−3.05	1	14
Methyl linoleate	$C_{19}H_{34}O_2$	294.47	5.74	2	1	−3.25	1	15

MF- Molecular formula; MW- Molecular weight; NA- Number of hydrogen bond acceptor; HD- Number of hydrogen bond donator; LV- Lipinski violation count; RB– Number of rotatable bonds; Log- Logarithmic skin permeation coefficient (Kp(cm/s); Log P- Octanol-water partition coefficient (Log P o/w).

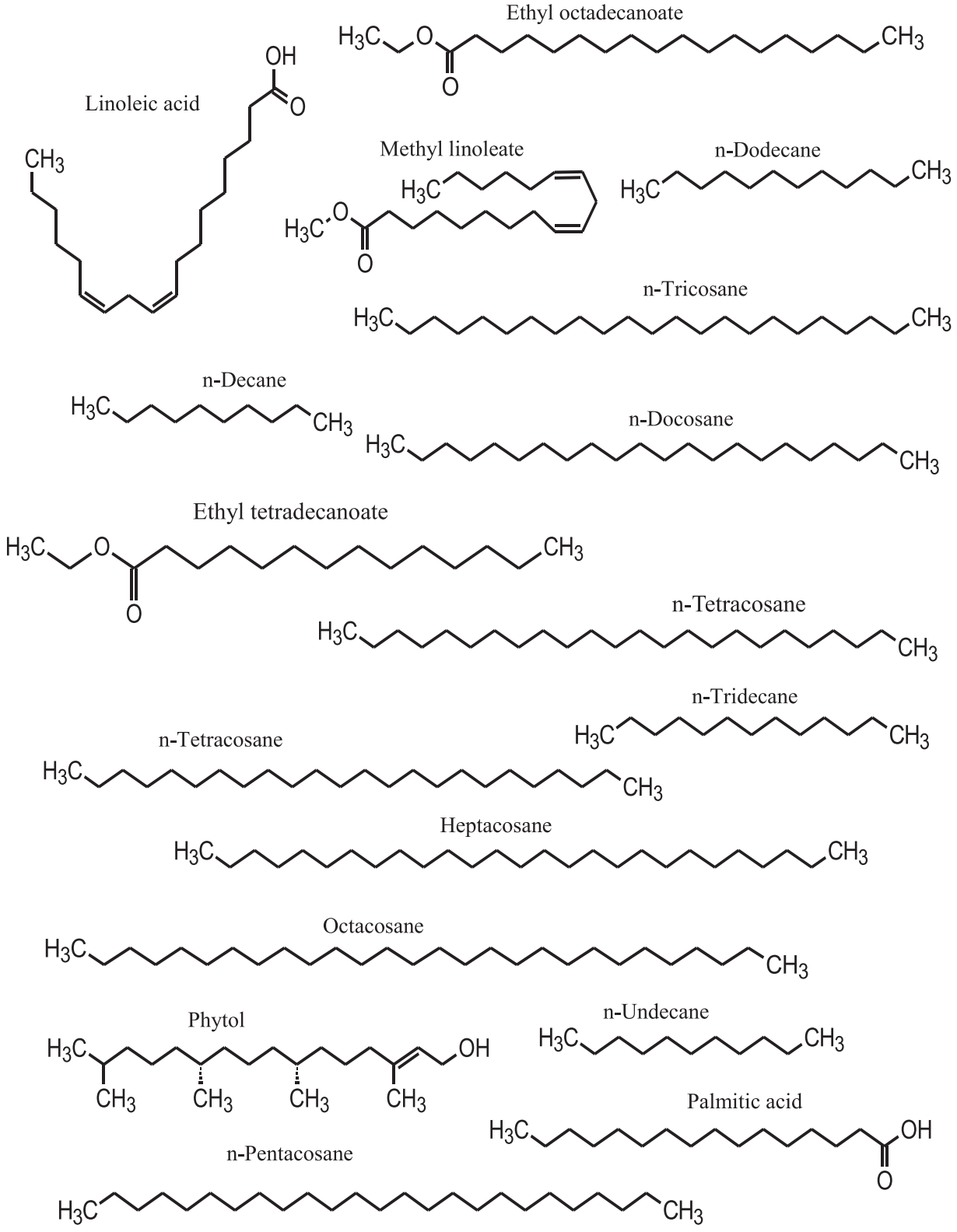

Fig. 1. Chemical structure of major compounds identified in the hydroalcoholic leaf extract of the *C. coromandelicum*.

2.5. *Tyrosinase inhibition (anti-aging) effect of* C. coromandelicum *leaf extract*

The tyrosinase inhibition potential of *C. coromandelicum* leaf extract was evaluated according to the methodology of Raj et al. (2023). Hundred μL of *C. coromandelicum* leaf extract (at different concentrations) and 70 μL of 4 mM L-DOPA solution (20 mM of phosphate buffer at a pH of 6.8) were mixed and kept for incubation (15 min) at 30 °C. The reaction was initiated by adding 20 μL tyrosinase enzyme and again incubated for 30 and 60 min at 37 °C. The inhibitory activity of samples was measured by reading the absorbance at 490 nm using kojic acid as a positive control. The inhibition

level of tyrosinase was estimated as given below:

$$\% \text{ of inhibition of sample} = \frac{\text{Control OD} - \text{Sample OD}}{\text{Control OD}} \times 100$$

2.6. Lipinski rule and the analysis of ADME/T properties of phytocompounds

To be active as a drug component, a promising ligand molecule/compound must reach its target with sufficient level of concentration, and must be stable in bioactive form for long duration for predictable level of biologic events that must occur inside the body of host. Swiss ADME web tool was used for the study of ADME prediction via *in silico* method hence, it is considered as a rapid and strong model to analyze physicochemical properties of a target compound, along with its pharmacokinetics as well as drug-likeness (Khadse et al., 2020).

2.7. Docking analysis of hydroalcoholic extract of C. coromandelicum leaf

Docking simulations used in the study were analysed using the software, Molecular Operating Environment (Ver. 2015.10) as per the methodology of Shah et al. (2021). The 3D crystal structure of selected proteins for α-amylase (PDB ID: 7TAA), α-glucosidase (PDB ID: 3WY1), COX-2 (PDB ID: 4COX), and tyrosinase (PDB ID: TY9X) enzymes were retrieved from the RCSB-Protein Databank (www.rcsb.org). Initially, water molecule and heteroatoms were removed from the selected proteins. Then, to the protein molecules, polar hydrogen atoms were added and their potential were also fixed. The receptor binding site of protein was identified using the London dG and GBVI/WSA dG default functions in the MOE software. Energy minimization process was optimized under MMFF94 force field. Effective ligand was conformed through the values of minimum docking scores or interaction energy estimation of enzyme-ligand complex system along with binding interaction.

2.8. Statistical analysis

All the obtained results of were expressed as mean±STD (n = 3) and evaluated using ANOVA with the Tukey comparison test. P value of <0.05 regarded as statistically significant. GraphPad Prism version 9.3 was used for the graphical representation of obtained data.

3. Results

3.1. In vitro hypoglycemic, anti-inflammatory, and anti-aging effects of C. coromandelicum leaf

The hydroalcoholic extract of *C. coromandelicum* leaf showed the extraction yield of 11.09% and found to be effective against α-amylase enzyme with inhibition percentage of 86.09±0.38 at 250 μg/mL which was comparable to the standard acarbose (25.55 ± 0.45 μg/mL) that showed 90.90±0.37% of inhibition (Fig. 2A) with an IC_{50} value of 42.51 ± 0.51 μg/mL. The inhibition effect of *C. coromandelicum* leaf extract against α-glucosidase enzyme was recorded as 83.53 ± 0.27% at 250 μg/mL (Fig. 2B) with an IC_{50} value of 30.82 ± 1.33 μg/mL. Likewise, hydroalcoholic leaf extract showed highest bovine serum albumin denaturation efficacy with the inhibition percentage of 73.17 ± 0.13 at 250 μg/mL with an IC_{50} value of 45.40 ± 1.75 μg/mL. The results are comparable with those of diclofenac which showed an IC_{50} value of 41.15 ± 0.45 μg/mL with inhibition percentage of 79.84 ± 0.26 at 250 μg/mL (Fig. 2C, Table S1).

Anti-aging potential of the hydroalcoholic leaf extract by tyrosinase inhibition assay showed significant inhibition of 76.78 ± 0.08% at a concentration of 250 μg/mL with an IC_{50} value of 56.92 ± 0.72 μg/mL (Fig. 2D). It was also comparable to the standard, kojic acid which showed 77.50 ± 0.21% of inhibition and IC_{50} value of 42.99 ± 0.36 μg/mL at the same concentration used for the plant extract. In all the assays (except for tyrosinase inhibition assay), the lower concentrations of plant extract showed significant difference ($p \leq 0.1$) with the standards used in the study. In the α-amylase enzyme inhibition assay, all the concentrations showed significantly different values ($p \leq 0.1$) except for 150 μg/mL, which showed significance of $p \leq 0.01$. Whereas, 150 and 200 μg/mL concentration in α-glucosidase enzyme inhibition assay and 50 μg/mL in tyrosinase inhibition assay displayed no significant different with the standards (Fig. 2).

3.2. ADME prediction and physicochemical properties of selected compounds

The best docked ligand molecules with their binding energy, hydrogen bond natures, and their corresponding amino acid residues were explored. The Lipinski's rule has been used to assess drug-likeness of compounds or ligands of the hydroalcoholic leaf extract of *C. coromandelicum*. Molecular properties such as logarithmic skin permeation coefficient, number of rotatable bonds, coefficient of octanol-water partition (Log P), molecular weight of compounds, nature of hydrogen bond acceptors, and hydrogen bond donors for selected ligands recorded (Table 1). The ligand 9 (octacosane) and 4 (heptacosane) showed high rotatable bonds while ligand 12 (n-undecane) and 13 (n-decane) exhibited low rotatable bonds.

The docking analysis of 7TAA (α-amylase enzyme) with ligands to identify specific amino acid residues was found to be composed of Arg204, Arg344, Asp340, Gln35, His80, Glu230, Trp83, Tyr75, Tyr82, Tyr256 and Tyr355 (Table S2 and Fig. 3). The docking score and binding interactions of ligands with α-glucosidase enzyme (3WY1) are presented in Table S3. The binding score of the ligand compounds ranged from −7.54 to 4.89 kcal/mol. Ligand 15 (phytol) (Fig. 4A) showed best interactions and docking score (−7.54 kcal/mol) with one conventional H-bond and interaction of two amino acids (Thr445, His348). Similarly, ligand 14 (ethyl tetradecanoate) (Fig. 4B) and ligand 1 (palmitic acid) (Fig. 4C) showed best docking score and each ligand binding with two amino acid residues −6.98 kcal/mol (Lys352 and Thr445) and −6.41 kcal/mol (Gln531 and Phe516), respectively. It was further observed that, ligands 16 (linoleic acid; Fig. 4D) and 17 (methyl linoleate) showed strong interaction with α-glucosidase enzyme with the docking score of −6.94 and −6.90 kcal/mol, respectively.

Table S4 shows the binding interaction of compounds with 4COX protein. The best finding poses (Glu-B46, Lys-A546, and Lys-B137) and docking scores (−7.46 kcal/mol) were obtained for ligand 16 (linoleic acid) (Fig. 5A). The ligand 2 (ethyl octadecanoate) (Fig. 5B) showed the best binding affinity with a notable docking score (−7.41 kcal/mol) similar to that of ligand 17 (methyl linoleate −6.03 kcal/mol; Fig. 5C). The ligand 1 (palmitic acid) (Fig. 5D) showed two hydrogen bond interactions (Arg-B44 and Asp-B125) with best docking score of −6.49 kcal/mol. The docking interaction of tyrosinase (2Y9X) protein with bioactive compounds are given in Table S5. The binding affinity values varied from −8.13 to −5.15 kcal/mol against the target protein, 2Y9X. The ligand 15 (phytol) with 2Y9X interaction has one hydrogen bond interaction with −6.61 kcal/mol as docking score (Fig. 6A). Similarly, ligand 16 (linoleic acid) showed best interaction (−6.57 kcal/mol) in 2Y9X protein with one hydrogen bond (Tyr236) interactions (Fig. 6B). Besides, ligand 17 (methyl linoleate) showed notable binding score (−6.02 kcal/mol) with one hydrogen bond interactions (Leu11) at distance of 3.01 Å (Fig. 5C).

4. Discussion

The hydroalcoholic extract of *C. coromandelicum* was found to possess long-chain saturated and unsaturated hydrocarbons which

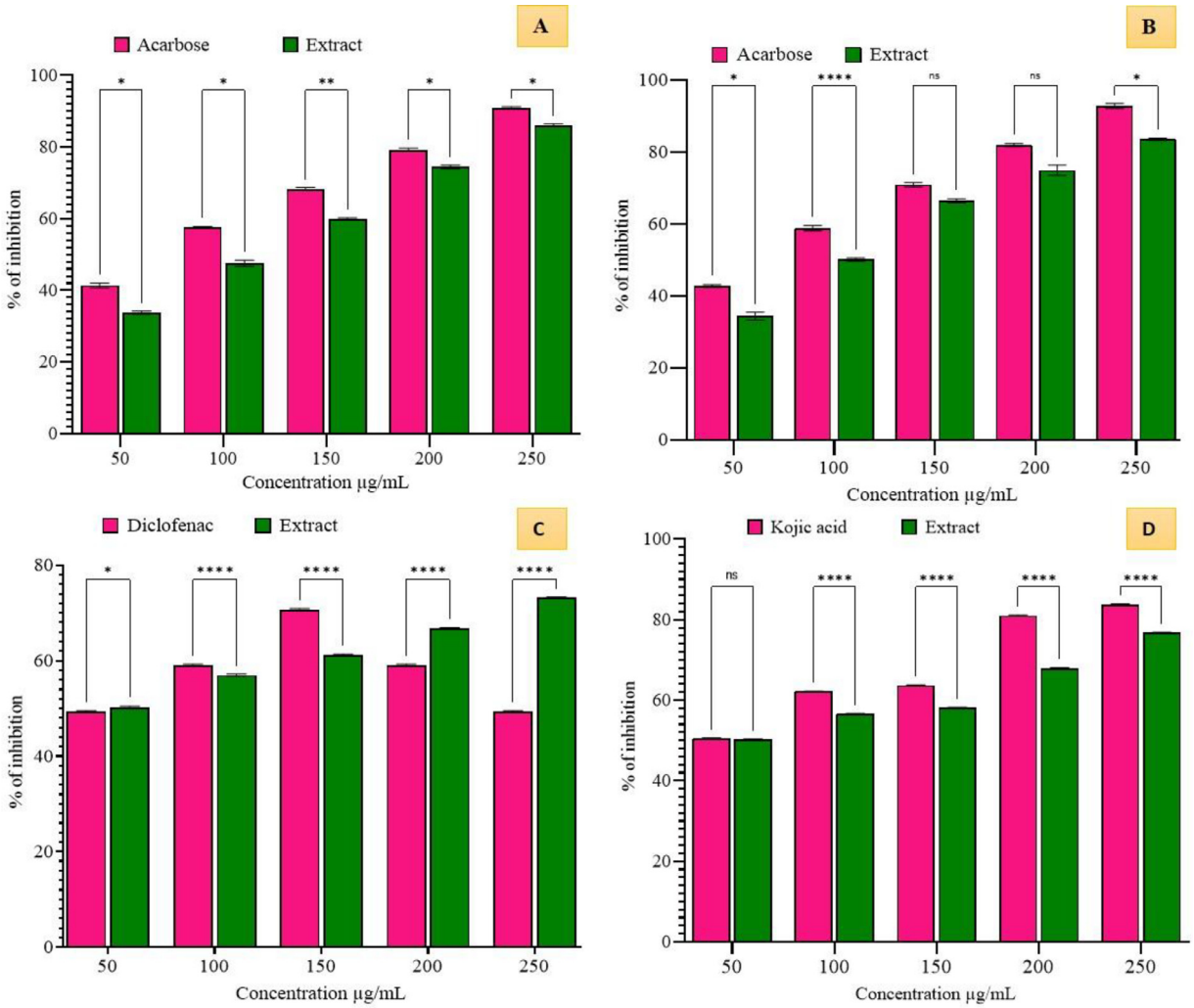

Fig. 2. Enzyme inhibition effect of *C. coromandelicum* leaf extracts. A - α-amylase enzyme inhibition assay; B - α-glucosidese enzyme inhibition assay; C – anti-inflammatory activity; D – tyrosinase enzyme inhibition activity. Different letters as superscripts in the figure are significant different values (*- $p \leq 0.1$; **- $p \leq 0.01$; *** - $p \leq 0.001$; **** - $p \leq 0.0001$; ns - no significance) compared to the standards, acarbose, diclofenac, and kojic acid.

are believed to be responsible for potent antidiabetic, anti-aging and anti-inflammatory activities. In a recent study conducted at our laboratory, an extract from *Pisonia grandis* R.Br. (family Nyctaginaceae) leaves with similar phytochemistry was shown to possess potent antioxidant, α-amylase, and α-glucosidase inhibitory effects (Gangapriya et al., 2022). The plant has been previously studied for its antioxidant, antibacterial and antidiabetic effects based on its usage against various ethnomedicinal applications (Amalraj et al., 2021). However, there is a lacuna in other enzymatic studies for anti-inflammatory and tyrosinase inhibitory activities.

Previous research showed that polyphenolic compounds, phenolic acids, flavonoids and tannins are characterised by varied inhibitory properties against the enzymes, α-glucosidase and α-amylase. In addition to the antioxidant effects, dietary polyphenolic compounds exhibited significant antidiabetic effects by inhibition of digestive enzymes (Ofosu et al., 2021). Kala et al. (2014) reported that antidiabetic activity of callus extract of *Canthium parviflorum* Lam. leaf showed α-glucosidase enzyme inhibition activity with an IC_{50} value of 904.52 μg/mL. As reported in the present study, ethanolic extract of *Nectandra angustifolia* (Schrad.) Nees & Mart. (family Lauraceae) leaf showed membrane stabilization effect on human red blood and protein denaturation activity in a dose-dependent manner (Ferrini et al., 2021) and n-butanol extract of *Hyacinthoides lingulata* (Poir.) Rothm. (family Asparagaceae) exhibited remarkable anti-inflammatory activity (84.82%) at a concentration of 200 μg/mL (Hanfer et al., 2022). The plant-derived compounds such as resveratrol, curcumin, colchicine and quercetin are characterised by excellent anti-inflammatory activity (Akhter et al., 2022).

In support of our research findings, the anti-aging potential of the compound 3,5-dihydroxy-4′,7-dimethoxyflavone obtained from the *Alpinia nigra* (Gaertn.) Burtt (family Zingiberaceae) leaf extract showed potential tyrosinase inhibition activity (68%) at a concentration of 60 μM (Gupta et al., 2021) and ethanolic extract of *Rosa chinensis* Jacq. (family Rosaceae) leaf showed strong tyrosinase enzyme inhibition activity with an inhibition range of 44 to 56% (Li et al., 2021). Our results were also in agreement with findings obtained by Peng et al. (2022) which showed positive correlation between the antioxidant and anti-tyrosinase effects of the plant products and their derivatives.

In silico ADMET studies are used to predict the pharmacokinetic and pharmaco-dynamic properties of a compound and to identify potential risks associated with its use. In the present study, binding affinity of selected compounds or ligands were investigated using molecular docking and the results showed that all the docked compounds had significant high binding affinity for the 7TAA protein. Heptacosane had the highest binding affinity, followed by linoleic acid, ethyl octadecenoate and phytol and highest docking score was observed for heptacosane, octacosane, ethyl octadecanoate, and n-

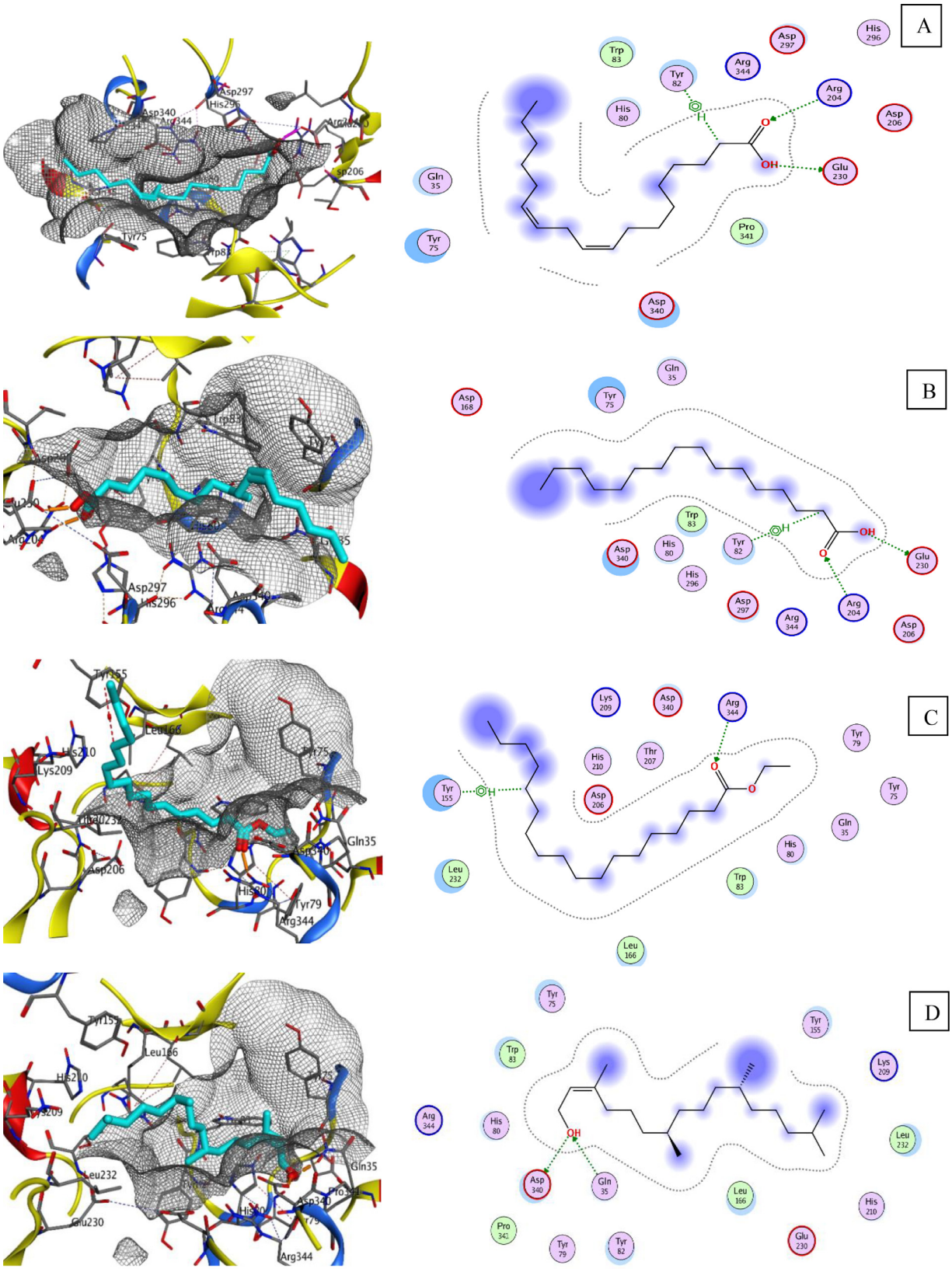

Fig. 3. Binding interaction of compounds with active site of α-amylase (7TTA). A - linoleic acid; B - palmitic acid; C - ethyl octadecanoate; D - phytol.

pentacosane with 7TAA. Also, the best docking interactions were observed for ethyl octadecanoate (−6.39 kcal/mol) and phytol (−6.10 kcal/mol). Palmitic acid, one of a saturated fatty acid which was considered as an essential compound for human body which had a key role in treating cancer, inflammation, cardiovascular and neuro-degenerative diseases (Wang et al., 2022).

Linoleic acid is considered as a good inhibitor of the enzyme α-glucosidase (3A47) that is bounded to have active site through

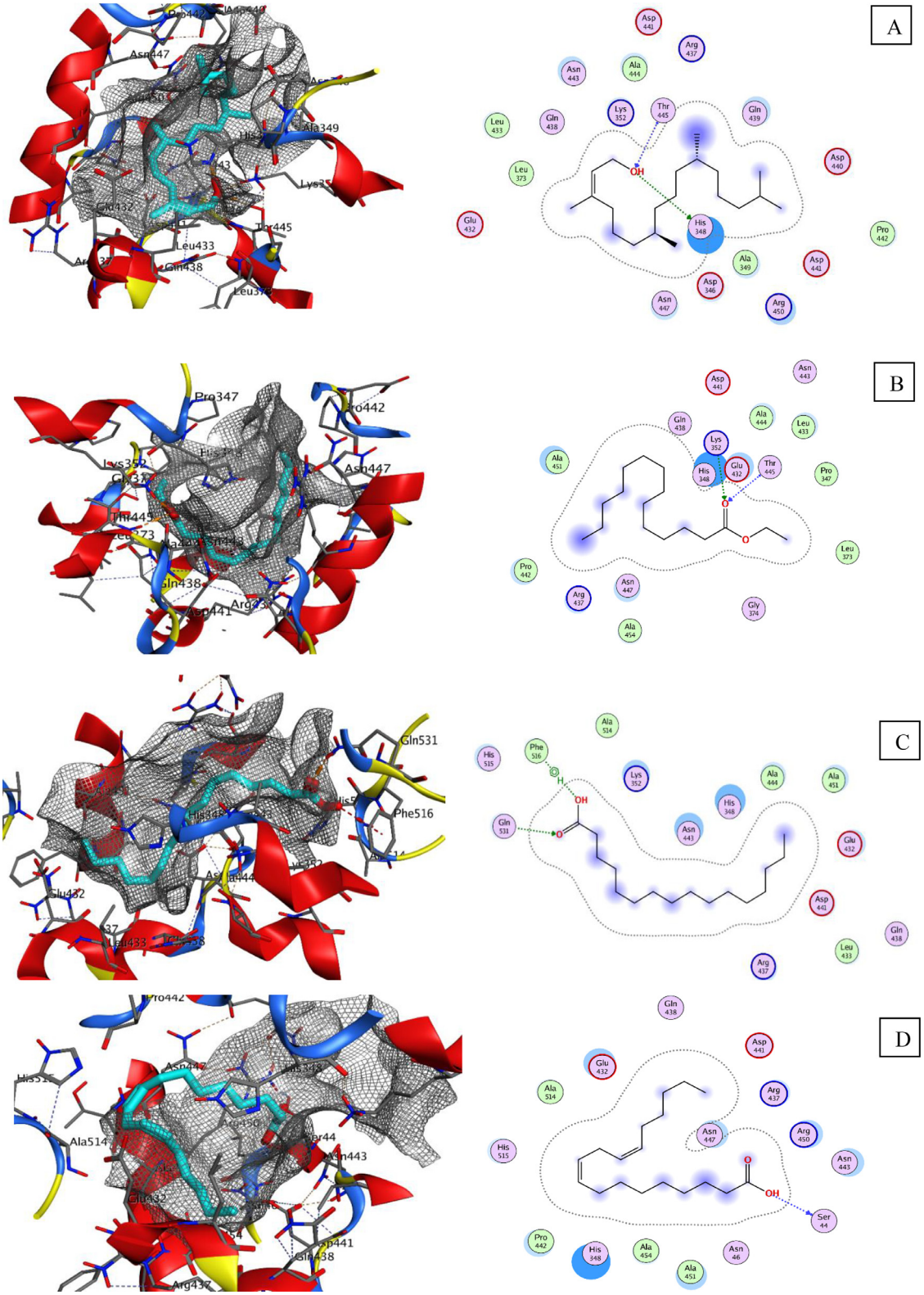

Fig. 4. Binding interaction of compounds with active site of α-glucosidase (3WY1). A - phytol; linoleic acid; B - ethyl tetradecanoate; C - palmitic acid; D - linoleic acid.

multiple interactions with the docking score of −5.89 kcal/mol (Shah et al., 2021). Binding energy data computed via docking studies revealed that all phytocompounds identified in hydroalcoholic extract of *C. coromandelicum* can potentially inhibit the enzyme activity of α-amylases and α-glucosidases. Interaction analysis of best docked compounds such as phytol, linoleic acid, palmitic acid and ethyl tetradecanoate exposed several hydrophobic bonds on the enzymes, α-amylases and α-glucosidases. Similarly, phytol and linoleic acid were well bound with α-glucosidase enzyme (docking score of −7.0629 and −7.1746 kcal/mol, respectively) (Mgbeje and Abu, 2020) which was in agreement with the results presented in this study.

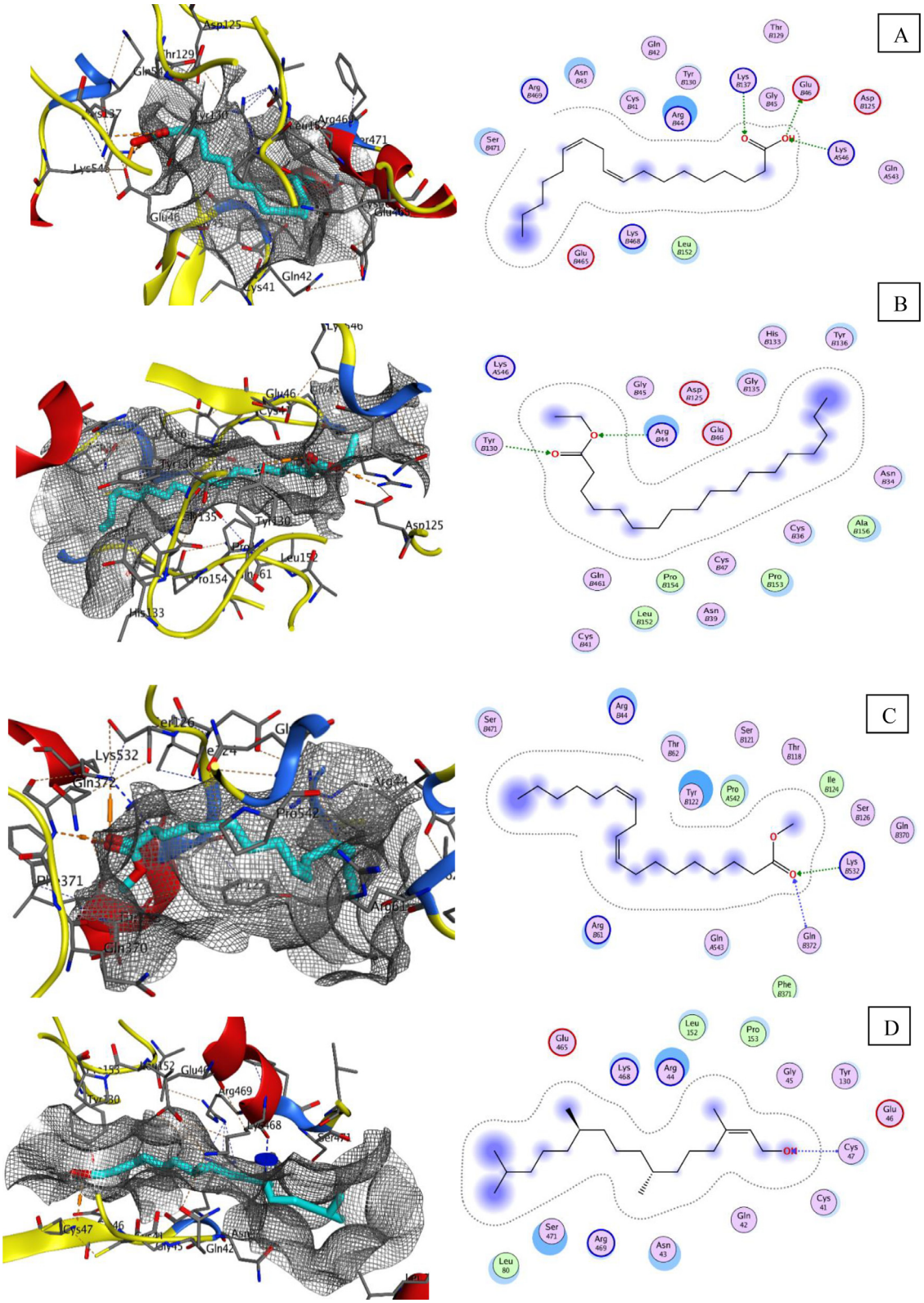

Fig. 5. Binding interaction of compounds with active site of cyclooxygenase-2 enzyme (4COX). A - linoleic acid; B - ethyl octadecanoate; C - methyl linoleate; D - phytol.

The metabolites like curcumin, resveratrol, quercetin, and green tea catechins bind to and inhibit the activity of COX enzyme which regulates the production of inflammatory mediators and prostaglandins (Nigussie et al., 2021). To understand the working principle of these compounds, binding pattern of COX using molecular docking can be done by predicting the binding orientation and affinity of a ligand molecule to a protein receptor (Labib et al., 2017). As seen in docking results of compounds against 4COX (Table S4), all the cyclic compounds bind to 4COX and this is consistent with their anti-inflammatory activity. Previous studies revealed that linoleic acid possess antioxidant, hepatoprotective, antiarthritic and anti-inflammatory activities (Alminderej et al., 2020). Linoleic acid also possesses

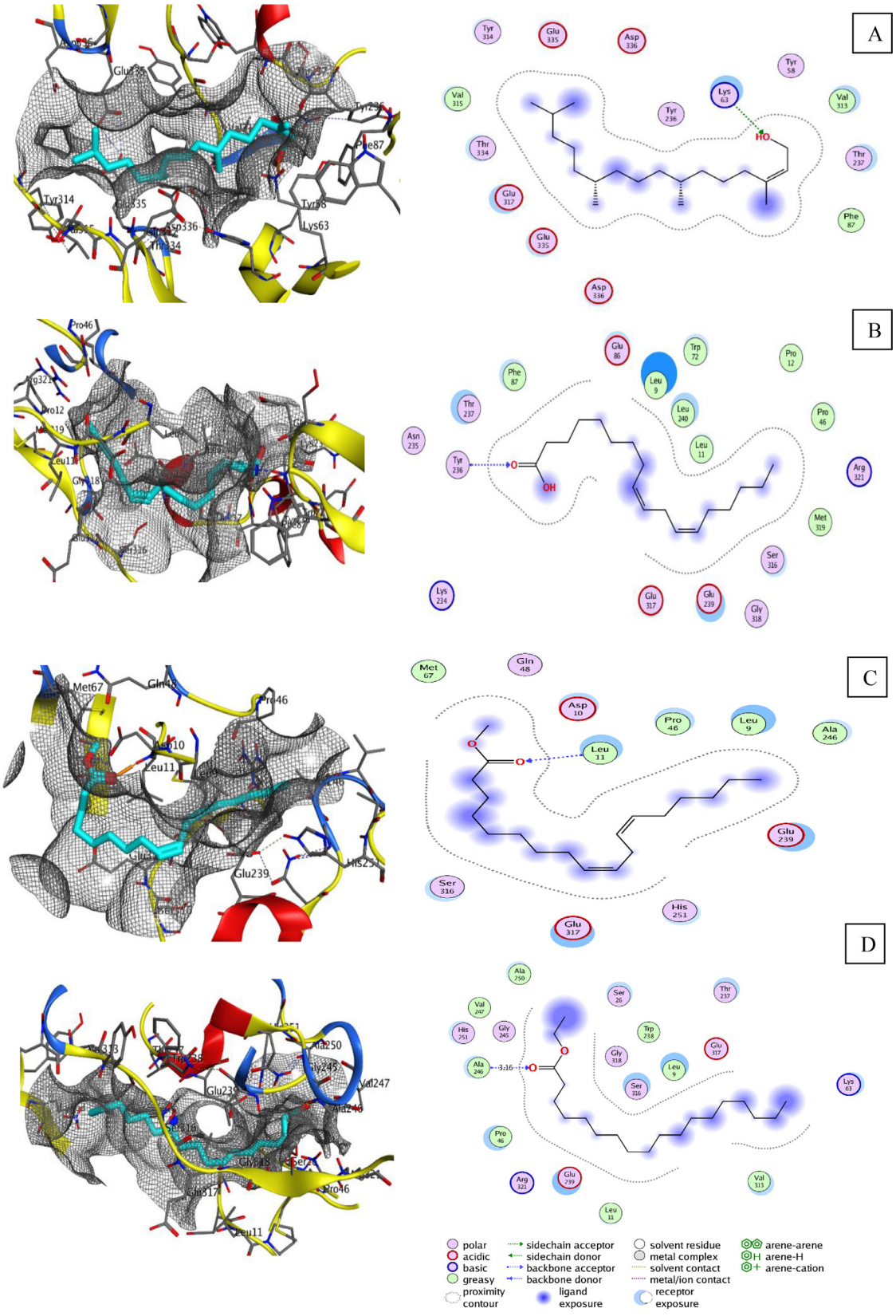

Fig. 6. Binding interaction of compounds with active site of tyrosinase enzyme (2Y9X). A - phytol; B - linoleic acid; C - ethyl octadecanoate; D - methyl linoleate.

antidiabetic activity by reducing insulin resistance (Yoon et al., 2021) and exhibits anti-inflammatory activity by suppressing the expression of the NF-κB subunit p50 and restoring PPARα *in vitro* RAW 264.7 cell studies (Saiki et al., 2017).

Ligand 15 (phytol) also displayed good affinity towards the enzyme active site (docking score of −6.83 kcal/mol). Previously, an acyclic diterpene phytol isolated from the hemp (*Cannabis sativa* L., family Cannabaceae) seed oil demonstrated anti-inflammatory

activity in human monocyte-macrophages (Claro-Cala et al., 2022). In a recent study, anti-inflammatory activity of the bioactive constituents from *Piper cubeba* L.f. (family Piperaceae) through *in silico* analysis revealed that, β-caryophyllene oxide (docking score of −5.8 kcal/mol), α-selinene (docking score of −5.4 kcal/mol) and viridiflorol (docking score of −5.1 kcal/mol) were docked well with the PTP1B (PDB ID: 1HD2) (Alminderej et al., 2020). Likewise, palmitic acid (−60.28), 1-octadecanol (−55.36 kcal/mol), myristic acid (−54.46 kcal/mol) and caryophyllene oxide (−34.87 kcal/mol) isolated from *Pinus roxburghii* Sarg. (family Pinaceae) showed good binding energy with human glucocorticoid receptor (PDB ID: 1M2Z) (Labib et al., 2017).

In support of the current research findings, the polyphenolic compounds obtained from the plants showed good docking score (−3.4 to 7.9 kcal/mol) and interaction (Asp312, Glu335, Glu356, Lys379) with tyrosinase protein (2Y9X) (Sohretoglu et al., 2018). An isoflavone, 5,7-dihydroxy-4-methoxy-3−3-methyl-2-hydroxybuten-3-yl) isolated from the fruits of *Ficus hispida* L.f. (family Moraceae) showing good docking (−4.53 kcal/mol) score with tyrosinase (2Y9X) and interacted well with several active site amino acid residues including MET280, HIS259, HIS263, PHE264, and ARG268 by non-covalent interaction (Cheng et al., 2021). This result suggests that the hydroalcoholic extract of *C. coromandelicum* leaf could be used as treatment for hyperpigmentation disorders like melisma.

Chronic inflammation has been linked to an increased risk of diabetes while aging is linked to increased risk of both diabetes and inflammation (Li et al., 2022). Diabetes is also associated with an increased risk of certain kinds of cancer, which can be triggered by inflammation. Aging contributes to an increased risk of diabetes, inflammation, and other age-related disorders including Alzheimer's and Parkinson's diseases. Moreover, inflammation may raise the likelihood of developing cardiovascular diseases which is a key risk factor for diabetes and related diseases (Muriach et al., 2014; Guarner and Rubio-Ruiz, 2015). So, there is need for an ideal and effective drug in treating diabetes, inflammation and anti-aging complications that would reduce the need for multiple medications and eventually simplify the treatment process.

5. Conclusion

Molecular docking analysis revealed better binding affinity to human pancreatic enzymes, tyrosinase enzyme and cyclooxygenase-2. *In silico* molecular docking analysis of the identified compounds from *C. coromandelicum* leaf showed good binding score, ranging from - 8.59 Kcal/mol to - 4.43 Kcal/ mol with human pancreatic α-amylase (7TAA) and −7.32 Kcal/mol to −4.09 Kcal/ mol with α-amylase enzyme (3BAJ) as target proteins. The compounds linoleic acid, phytol, methyl linoleate, palmitic acid, and ethyl tetradecanoate showed better interactions with low docking score and high binding ability with α-amylase, α-glucosidase, COX-2, and tyrosinase target proteins when compared with other compounds. The results obtained in the study could be useful towards designing new molecules and pharmacophore-based designing of potential multi-targeted compounds with potential interactions and efficacy against diabetes and inflammation related diseases. The study also examined the effects of leaf extracts on pancreatic β-cells on molecular pathways in the regulation of glucose homeostasis, to protect against oxidative stress, and to modulate the expression of inflammatory mediators. Further comprehensive research is warranted to elucidate the mechanism of action of identified phytochemical compounds based on their pharmacological properties recorded in this study.

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

CRediT authorship contribution statement

Singamoorthy Amalraj: Conceptualization, Data curation, Investigation, Methodology, Writing – original draft. **Shailendra S. Gurav:** Data curation, Formal analysis, Writing – review & editing. **Mohan G Kalaskar:** Data curation, Formal analysis, Writing – review & editing. **Alfred Maroyi:** Data curation, Formal analysis, Writing – review & editing. **Muniappan Ayyanar:** Investigation, Methodology, Funding acquisition, Writing – review & editing.

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Supplementary materials

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References

Akhter, S., Hossain, M.W., Sultana, S., Jharna, Jannatul Ferdous, Meghla, Nigar Sultana, Alam, R., AnisUlHaque, K., Rahman, M.M., 2022. *Ruellia prostrata* Poir. activity evaluated by phytoconstituents, antioxidant, antiinflammatory, antibacterial activity, and *in silico* molecular functions. J. Saudi Chem. Soc. 26, 101401.

Ali, A., Khandelwal, S., Panja, S., Majumder, P., Dutta, A., 2022. Oxygen reduction reaction in nature and its importance in life. Oxygen Reduction Reaction. Elsevier, pp. 1–43.

Alminderej, F., Bakari, S., Almundarij, T.I., Snoussi, M., Aouadi, K., Kadri, A., 2020. Antioxidant activities of a new chemotype of *Piper cubeba* fruit essential oil (Methyleugenol /Eugenol): *in silico* molecular docking and ADMET studies. Plants 9, 1534.

Amalraj, S., Krupa, J., Sriramavaratharajan, V., Mariyammal, V., Murugan, R., Ayyanar, M., 2021. Chemical characterization, antioxidant, antibacterial and enzyme inhibitory properties of *Canthium coromandelicum*, a valuable source for bioactive compounds. J. Pharm. Biomed. Anal. 192, 113620.

Batool, M., Ahmad, B., Choi, S., 2019. A structure based drug discovery paradigm. Int. J. Mol. Sci. 20, 2783.

Cheng, J.X., Li, Y.Q., Cai, J., Zhang, C.F., Akihisa, T., Li, W., Kikuchi, T., Liu, W.Y., Feng, F., Zhang, J., 2021. Phenolic compounds from *Ficus hispida* Lf as tyrosinase and melanin inhibitors: biological evaluation, molecular docking, and molecular dynamics. J. Mol. Struct. 1244, 130951.

Cisneros-Yupanqui, M., Lante, A., Mihaylova, D., Krastanov, A.I., Rizzi, C., 2023. The α-amylase and α-glucosidase inhibition capacity of grape pomace: a review. Food Bioproc. Technol. 16 (4), 691–703.

Claro-Cala, C.M., Grao-Cruces, E., Toscano, R., Millan-Linares, M.C., Montserrat-de la Paz, S., Martin, M.E., 2022. Acyclic diterpene phytol from hemp seed oil (*Cannabis sativa* L.) exerts anti-inflammatory activity on primary human monocytes-macrophages. Foods 11 (15), 2366.

Dwivedi, P.S.R., Patil, R., Khanal, P., Gurav, N.S., Murade, V.D., Hase, D.P., Kalaskar, M.G., Ayyanar, M., Chikhale, R.V., Gurav, S.S., 2021. Exploring the therapeutic mechanisms of *Cassia glauca* in diabetes mellitus through network pharmacology, molecular docking and molecular dynamics. RSC Adv. 11 (62), 39362–39375.

Ferrini, L., Rodríguez, J.P., Pablo, J., Olea, G., Ojeda, G.A., Ricciardi, G., Torres, A.M., Aguirre, M.V., 2021. Antiinflammatory activity of *Nectandra angustifolia* (*Laurel Amarillo*) ethanolic extract. J. Ethnopharmacol. 272, 113937.

Gangapriya, P., Arulraj, M.S., Amalraj, S., Murugan, R., Ayyanar, M., 2022. Phytochemical composition, enzyme inhibitory potential, antioxidant and antibacterial activities of *Pisonia grandis* R. Br. (lettuce tree) leaves. J. Food Meas. Charact. 16 (4), 2864–2874.

Guarner, V., Rubio-Ruiz, M.E., 2015. Low-grade systemic inflammation connects aging, metabolic syndrome and cardiovascular disease. Aging and health-A systems biology perspective. Interdiscip. Top. Gerontol. Geriatr. 40, 99–106.

Gupta, M.K., Senthilkumar, S., Chiranjivi, A.K., Banik, K., Girisa, S., Kunnumakkara, A.B., Dubey, V.K., Rangan, L., 2021. Antioxidant, anti-tyrosinase and anti-inflammatory activities of 3, 5-dihydroxy-4′, 7-dimethoxyflavone isolated from the leaves of *Alpinia nigra*. Phytomed. Plus. 1 (3), 100097.

Hanfer, M., Benramdane, Z., Cheriet, T., Sarri, D., Menad, A., Mancini, I., Seghiri, R., Ameddah, S., 2022. Chemical constituents, *in vitro* antiinflammatory, antioxidant

and hemostatic activities of the nbutanol extract of *Hyacinthoides lingulata* (Poir.) Rothm. Nat. Prod. Res. 36, 3124–3128.
Henein, M.Y., Vancheri, S., Longo, G., Vancheri, F., 2022. The role of inflammation in cardiovascular disease. Int. J. Biol. Sci. 23 (21), 12906.
Kala, S.C., Mallikarjuna, K., Reddy, C.S., 2014. *In vitro* pharmacological investigations of using leaf callus extracts of *Canthium parviflorum* Lam. Br. Biotechnol. J. 4, 325–338.
Khadse, S.C., Amnerkar, N.D., Dighole, K.S., Dhote, A.M., Patil, V.R., Lokwani, D.K., Ugale, V.G., Charbe, N.B., Chatpalliwar, V.A., 2020. Hetero-substituted sulfonamido-benzamide hybrids as glucokinase activators: design, synthesis, molecular docking and *in silico* ADME evaluation. J. Mol. Struct. 1222, 128916.
Khan, H., Sharma, K., Kumar, A., Kaur, A., Singh, T.G., 2022. Therapeutic implications of cyclooxygenase (COX) inhibitors in ischemic injury. J. Inflamm. Res. 71, 277–292.
Khanal, P., Chikhale, R., Dey, Y.N., Pasha, I., Chand, S., Gurav, N., Ayyanar, M., Patil, B., Gurav, S., 2022. Withanolides from *Withania somnifera* as an immunity booster and their therapeutic options against COVID19. J. Biomol. Struct. Dyn. 40, 5295–5308.
Labib, R.M., Youssef, F.S., Ashour, M.L., AbdelDaim, M.M., Ross, S.A., 2017. Chemical composition of *Pinus roxburghii* bark volatile oil and validation of its antiinflammatory activity using molecular modelling and bleomycin induced inflammation in albino mice. Molecules 22, 1384.
Li, H., Meng, Y., He, S., Tan, X., Zhang, Y., Zhang, X., Wang, L., Zheng, W., 2022. Macrophages, chronic inflammation, and insulin resistance. Cells 11 (19), 3001.
Li, M., Xie, J., Bai, X., Du, Z., 2021. Antiaging potential, antityrosinase and antibacterial activities of extracts and compounds isolated from *Rosa chinensis* cv. "JinBian.". Ind. Crops Prod. 159, 113059.
Mahishi, P., Srinivasa, B., Shivanna, M., 2005. Medicinal plant wealth of local communities in some villages in Shimoga District of Karnataka, India. J. Ethnopharmacol. 98, 307–312.
Mgbeje, B.I., Abu, C., 2020. Chemical fingerprinting of *Nauclea latifolia*, an antidiabetic plant, using GCMS. J. Complement. Altern. Med. Res. 9, 25–34.
Mishra, S.K., Balendra, V., Esposto, J., Obaid, A.A., Maccioni, R.B., Jha, N.K., Perry, G., Moustafa, M., Al-Shehri, M., Singh, M.P., Khan, A.A., Vamanu, E., Singh, S.K., 2022. Therapeutic antiaging strategies. Biomedicines 10 (10), 2515.
Muriach, M., Flores-Bellver, M., Romero, F.J., Barcia, J.M., 2014. Diabetes and the brain: oxidative stress, inflammation, and autophagy. Oxid. Med. Cell. 2014, 102158.
Nigussie, D., Makonnen, E., Tufa, Takele Beyene, Brewster, M., Legesse, B.A., Fekadu, A., Davey, G., 2021. Systematic review of Ethiopian medicinal plants used for their antiinflammatory and wound healing activities. J. Ethnopharmacol. 276, 114179.
Ofosu, F.K., Elahi, F., Daliri, E.B., Tyagi, A., Chen, X.Q., Chelliah, R., Kim, J., Han, S., Oh, D., 2021. UHPLCESIQTOFMS/MS characterization, antioxidant and antidiabetic properties of sorghum grains. Food Chem. 337, 127788.
Peng, Z., Wang, G., Zeng, Q., Li, Y., Liu, H., Wang, J.J., Zhao, Y., 2022. A systematic review of synthetic tyrosinase inhibitors and their structureactivity relationship. Crit. Rev. Food Sci. Nutr. 62, 4053–4094.
Raj, S., Santhi, V., Amalraj, S., Murugan, R., Gangapriya, P., Pragadheesh, V., Sundaresan, V., Gurav, S., Paramaguru, P., Arulmozhian, R., 2023. A comparative analysis of leaf essential oil profile, *in vitro* biological properties and *in silico* studies of four Indian Guava (*Psidium guajava* L.) cultivars, a promising source of functional food. S. Afr. J. Bot. 153, 357–369.
Rana, A., Pathak, S., Lim, D.K., Kim, S.K., Srivastava, R., Sharma, S.N., Verma, R., 2023. Recent advancements in plant-and microbe-mediated synthesis of metal and metal oxide nanomaterials and their emerging antimicrobial applications. ACS Appl. Nano Mater 6 (10), 8106–8134.
Sapkal, P.R., Tatiya, A.U., Firke, S.D., Redasani, V.K., Gurav, S.S., Ayyanar, M., Jamkhande, P.G., Surana, S.J., Mutha, R.E., Kalaskar, M.G., 2023. Phytochemical profile, antioxidant, cytotoxic and antiinflammatory activities of stem bark extract and fractions of *Ailanthus excelsa* Roxb.: *in vitro, in vivo* and *in silico* approaches. Heliyon 9 (5), e15952.
Saiki, P., Kawano, Y., Van Griensven, L.J., Miyazaki, K., 2017. The anti-inflammatory effect of *Agaricus brasiliensis* is partly due to its linoleic acid content. Food Funct. 8, 4150–4158.
Seo, Y., Bang, S., Son, J., Kim, D., Jeong, Y., Kim, P., Yang, J., Eom, J., Choi, N., Kim, H.N., 2021. Brain physiome: a concept bridging *in vitro* 3D brain models and *in silico* models for predicting drug toxicity in the brain. Bioact. Mater. 13, 135–148.
Shah, M., Murad, W., Rehman, U., Mubin, S., AlSabahi, J.N., Ahmad, M., Zahoor, M., Ullah, O., Waqas, M., Ullah, S., 2021. GCMS analysis and biomedical therapy of oil from nhexane fraction of *Scutellaria edelbergii* Rech. f.: *in vitro, in vivo*, and *in silico* approach. Molecules 26, 7676.
Sohretoglu, D., Sari, S., Barut, B., Ozel, A., 2018. Tyrosinase inhibition by some flavonoids: inhibitory activity, mechanism by *in vitro* and *in silico* studies. Bioorg. Chem. 81, 168–174.
Wang, J., Li, H., Meng, X., Tong, P., Liu, X., 2022. Biosynthesis of c9, t11conjugated linoleic acid and the effect on characteristics in fermented soy milk. Food Chem. 368, 130866.
Yoon, S.Y., Ahn, D., Hwang, J.Y., Kang, M.J., Chung, S.J., 2021. Linoleic acid exerts antidiabetic effects by inhibiting protein tyrosine phosphatases associated with insulin resistance. J. Funct. Foods 83, 104532.