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Original Article

Investigating the α-Amylase Inhibitory Effects of *Adansonia digitata* L.: A Comprehensive *in vitro* and Computational Study

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ABSTRACT

Background: The study explores *Adansonia digitata* L.'s potential in diabetes management through phytochemical and protein analyses. **Materials and Methods:** Eight identified phytoconstituents, comprising alkaloids, terpenes and steroids, were predicted to modulate diabetic proteins. Network analysis revealed a significant role for compound a (Kaempferol-3-O-rutinoside) in interacting with key molecules. Structural evaluations of α-amylase (PDB: 4W93) using PROCHECK and Chi1-Chi2 highlighted protein quality. **Results:** The molecular docking study concluded that compound a (Kaempferol-3-O-rutinoside) showedα-amylase inhibition with-8.21 kcal/mol as compared to standard drug-7.81 kcal/molacarbose. Furthermore, ADMET predictions indicated some deviations from ideal oral bioavailability for compound a, emphasizing considerations for its pharmaceutical application. The analysis also confirmed the non-toxic nature of compound a, elucidating its safety profile. The hydro alcoholic plant extract demonstrated selective inhibition of α-amylase, with IC_{50} values of 33.90 µg/mL. The Ethyl Acetate Fraction (EAF) exhibited an IC_{50} value of 29.21 µg/mL at a concentration of 50 µg/mL, compared to the standard acarbose, which had an IC_{50} value of 23.18 µg/mL. **Conclusion:** Using *in vitro* activity and computer-aided drug design models, the current study revealed that *Adansonia digitata* L. possesses potential α-amylase inhibitory characteristics.

Keywords: Anti-diabetic, Gene Ontology, Kaempferol-3-O-rutinoside, Molecular Docking, Network pharmacology.

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INTRODUCTION

The wealth of herbal wisdom found in Ayurvedic and Unani medicine is highlighted in ancient literature. A nearly Indian medical book called Charaka Samhita (1000 B.C.) explores the wide use of more than 2000 plants for therapeutic purposes, capturing a wealth of information and advantages gained from using herbal treatments in our in our initial.[1] Among the most useful therapeutic herbs, the Baobab (*Adansonia digitata* L.) belongs to Family: Malvaceae, stands as an iconic, majestic sub-tropical tree indigenous to Africa, revered forits cultural significance as the "arbre a palabre," a communal space for elders to address issues.[2] Beyond its emblematic role, the Baobab has gained attention from pharmaceutical companies and researchers over the last decade due to its multifaceted traditional applications. Its roots delve into medicinal, nutritional and cosmetic realms, drawing interest for potential benefits. This botanical giant, found across numerous African countries, symbolizes a convergence of heritage and modern exploration, prompting a renaissance in the exploration of its diverse and valuable properties.[3] Medicinal herbs, with their natural reservoirs of medicinal components, offer valuable solutions for addressing chronic disorders, providing essential support for overcoming health challenges similar to Diabetes Mellitus (DM), is the chronic metabolic disease characterised by elevated blood sugar levels, which continues to pose a significant global health challenge.[4] As per the report from International Diabetes Federation (IDF), it is approximated that 537 million adults are living with diabetes and 6.7 million death shave been reported in 2021. Diabetes incidence is estimated to rise to 643 million by 2030 and 783 million by 2045.[5] One of the primary complications associated with diabetes deregulated carbohydrate metabolism, marked by elevated levels of postprandial glucose, further exacerbates diabetic complications. In this context, the development of multifunctional molecules possessing antioxidant properties and the ability to inhibit key enzymes involved in glucose metabolism, such as alpha amylase, holds immense therapeutic potential.[6]

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Experts were interested in research in g novel medications derived from plants or their derivatives inlight of the previous discoveries. The current research reports a method for evaluating *in silico* antidiabetic activity prediction against α-amylase enzyme and to carryout pharmacokinetic research and Molecular docking is utilized to forecast the possible binding mechanisms of phytochemicals. The method of discovering new drugs has been completely transformed by the incorporation of computer approaches like *in silico* simulations and molecular modelling. With the use of these techniques, scientists can forecast and examine molecule interactions at the atomic level, giving them vital information about the binding patterns and mechanisms of potential therapeutic candidates.

To closely study diabetes at a molecular level, molecular docking has proven to be an instrumental technique. Need for Molecular Docking in Diabetes Study like Identification of Drug Candidates, Mechanistic Insights and Optimization of Lead Compounds. Objectives of Molecular Docking in Diabetes Study Structure Prediction: Binding Affinity Assessment Identification of Key Interactions Virtual Screening: Reducing Oxidative Stress and Inflammation: Molecular docking is a game-changer in the field of diabetes research, making the drug discovery process more efficient and effective.

Adansonia digitata L. extracts have been traditionally utilized for various medicinal purposes, showcasing notable antioxidant, anti-diabetic and cardiotonic properties according to previous research. These beneficial activities are attributed to their high vitamin C content, along with being rich in polyphenols and calcium. Previous studies have successfully isolated polyphenols and vitamin C from different plant sources using various isolation techniques. These compounds were then evaluated through *in vitro* studies and molecular docking to determine their molecular mechanisms and binding capacities. These studies concluded that bioactive molecules like polyphenols and vitamin C effectively bind to receptors, facilitating the desired anti-diabetic actions. The interest to undertake this investigation is due to the fact that no detailed study regarding molecular mechanism of action and receptor binding capacity of active chemical constituents from *Adansonia digitata* L. for Cardioprotective, Anti-diabetic and Anti-oxidant activity. Also it is reported that antioxidant property of drugs may be responsible for prevention of cellular damage (cell membrane) and cure of many chronic disorders such as cardiovascular, cancer and diabetic etc.

Chemical composition

Analysis of *Adansonia digitata* L. highlights crucial phytochemicals (a-h) essential for α-amylase enzyme inhibition. Our prior reported LC-MS studies, detailed in Figure 1 (A), illuminate the chemistry of significant chemical components in Baobab. This exploration unveils the plant's potential in harnessing specific compounds vital for modulating α-amylase activity, contributing to its medicinal relevance.

MATERIALS AND METHODS

Preparation of Extract

The *Adansonia digitata* L. plant specimen was collected from village Khanderajuri, Tq.-Miraj, Dist.-Sangli, Maharshtra, India 10th Jan 2021, the herbarium sheet was prepared and submitted to by Botanical Survey of India, Western Regional Centre, Pune. The authentication certificate with Authentifiaction No.: No.BSI/WRC/100-1/Tech.2020/126 is attached here with.

Before being employed, Plant fruit part gathered, cleaned, drained, chopped, dried, combined and sieved. 150 g of powder were added to a 70:30 ethanol and water mixture and the mixture was left for 7 days with frequent stirring to create a crude extract. After filtering, the extract was put to a Soxhlet apparatus using the identical solvent solution and departed for three days, boiling constantly.

Flavonoid Fractionation Preparation

The hydroalcoholic extracts underwent successive liquid-liquid extraction using ethyl acetate. Then, Every fraction was gathered. and dried to obtain the fraction of ethyl acetate (Flavonoid Fraction/F.Fraction/EAF) under lowered pressure.[7]

In vitro α-amylase inhibition assay

The objective of the study was to ascertain how well hydroalcoholic extract along with flavonoid fraction inhibited the amylase enzyme. The samples' α-amylase inhibitory characteristics were assessed using the methodology detailed in (Poovitha *et al.*, 2016). Phosphate buffered saline with a pH of 6.9 and 0.1 M was used to dissolve α-amylase from the pig pancreas (3 units/mL). The samples ranging in concentration from 50 to 250 µg/mL were subjected to a 10-min pre-incubation with enzyme at 37°C.

In the incubation medium, substrate (0.1% starch) is added, the reaction was started. To halt the reaction after 10 min of incubation, 250 µL of Dinitro Salicylic (DNS) reagent (1% 3,5-dinitro salicylic acid, 2% phenl, 0.05% Na_2SO_3 and 1% NaOH in aqueous solution) was introduced. By immersing reaction mixture in boiling water for 10 min, the reaction was stopped. A 40% potassium sodium tartrate solution was then added in an amount of 250 µL. After being cooled in a cold-water bath to room temperature, the absorbance at 540 nm was measured.[8]

Using Acarbose as a positive control, the percentage of inhibition was calculated using the following formula: (OD of blank-OD of test/OD of Blank)*100. The outcomes were displayed in IC_{50} values.

Identification of phytocompounds and their target identification

Phytochemicals present in *Adansonia digitata* L. were selected by using an extensive literature survey and mining of public repositories such as Dr. Duke's D B and IMPATT. All the chosen compounds SMILES were acquired from the database of PubChem (.https://pubchem.ncbi.nlm.nih.gov) and searched for protein-based forecasting in DIGEP-Pred at a Probable activity (Pa)>0.5 and Probable inactivity (Pi) ratio.

Phytoconstituents and proteins implicated with diabetes are mined

Adansonia digitata L. phytoconstituents were methodically extracted from our previously published LC-MS research. A database containing all of the details regarding these phytoconstituents, including their kinds, SMILES and PubChem CID, was methodically constructed. During the databasecreation process, duplicate entries of phytoconstituents were carefully eliminated to assure data correctness. For every phytoconstituent, the PubChem Database provided the canonical SMILES and PubChemCID.[9] For predicting possible targets, SMILES data were examined using the Digip pred program.[10] At the same time, diabetes-related proteins were identified by consulting known targets listed in the Therapeutic Target Database (TTD).[11] After then, the Gene ID for every protein designated as a target for diabetes mellitus was obtained from the Uniport.[12]

Network construction

The STRING database was searched for a list of proteins that were regulated up- and down. The KEGG (Kyoto Encyclopaedia of Gene and Genomes) pathway route database was used to locate the regulated protein and its related pathways. After constructing the active components and their targets of interaction associated to diabetic mellitus, the Information was entered into the Cytoscape 3.7.2 program, which allowed the user to "visualise and analyse the network from high to low modulating." To identify the proper protein targets that *Adansonia digitata* L.'s phytoconstituents have modified.Cytoscape 3.7.2 used to designa target network, visually analysed and screened in order to investigate the connection between phytocompounds and protein targets of diabetes mellitus compound.[13,14]

Gene Ontology (GO) and Kyoto Encyclopaedia of Genes and Genomes (KEGG) Pathways: An Enrichment Analysis

SRP plot bioinformatic resources were used to examine Common target GO and KEGG enrichment analysis in order to ascertain the roles of *Adansonia digitata* L. in regulating DM responses.

Molecular docking studies on α-amylase

The Schrodinger's glide Extra Precision mode (XP) was taken into consideration for docking in order to determine the inhibitors' orientation and binding interaction in the α-amylase pocket.[15] To construct ligands, Schrodinger's suit's Ligprep panel was utilised. Energy minimisation was accomplished using the Optimised Potential for Liquid Simulation (OPLS3e). Torsional flexibility was applied in order to achieve a more harmonious position. The α-amylase's crystal structure coordinates (PDB ID: 4W93) were obtained from protein data bank PDB. (https://www.rcsb.org) by comparing the crystal structure to the initial binding conformation. The proteins were pre-processing, which involved deleting ligands and water molecules from the PDB dataset, allocating bond ordering and producing PKa values at PH7±2. To produce a stable structure for more research, Energy minimization was used to further optimise the protein utilising an OPLS3e force field.[16] The co-crystal ligand with acarbosete to designate the binding location inside the target receptor was used to produce the grid. The ligand binding affinity with receptors was determined in terms of docking score, hydrogen bonds and pi-pi interactions prior to docking using Glide program in Extra Precision (XP) mode.[17,18]

Prediction of Physicochemical Properties

Using MolSoft (https://www.molsoft.com/), the phytocompounds of *Adansonia digitata* L. (a-h) were predicted for the drug likeness score and the "Lipinski's rule of 5" principle. Similarly, the ADME profile of synthesized drugs was predicted using the Schrödinger Qikprop module.[19,20] Moreover, utilizing Cheminformatic tools available on the free web, We looked at the molecular volume correlation studies, the Mol Log P values and the Topological Polar Surface Area (TPSA) to determine the molecule's potential bioavailability. The degree of lipophilicity of a molecule is measured by its Mol Log P value; a hi(https://stoptox.mml.unc.edu/) gher value indicates a more lipophilic molecule. For compounds (a-h), the STopTox server identified the piece that would either raise or decrease acute oral toxicity.[21,22]

RESULTS AND DISCUSSION

In vitro α-amylase inhibition assay

Using acarbose as positive control drug, The research evaluated the inhibitory potential ofhydroalcoholic extract and flavonoid fraction against α-amylase enzymes. The results, which are shown in Figure 1 (B), showed both hydroalcoholic extract and EAF had significant inhibitory action against α-amylase, with the first one having a IC_{50} value of 33.90 µg/mL and the latter having an IC_{50} value of 29.21 µg/mL. These actions were measured against the reference acarbose, which has an IC_{50} of 23.18 µg/mL for α-amylase. Phytoconstituents and proteinsimplicated with diabetes are mined

Adansonia digitata L. was found to have 13 distinct phytoconstituents based on open-source data and our previously published LC-MS research; 8 (a-h) of these were predicted for modifying the diabetes protein molecules (Table 1). Alkaloids,

terpenes and steroids were identified as these phytoconstituents. In a similar vein, surface proteins and enzymes made up the bulk of the targeted diabetic protein molecules.

Enrichment analysis of phytoconstituents protein targets

A combined C-T and T-P network shows how phytochemicals, protein targets and pathways are related to one another. The C-T-P network has 104 edges and 21 nodes. 13 distinct pathways influenced by proteins connected to diabetes mellitus were identified through the use of gene set enrichment analysis. Four pathways were identified by peer analysis of the interaction between proteins using KEGG pathway analysis to the pathogenesis of diabetes mellitus. Notably, Table 1 shows that the process of digestion and produced the most gene sets with the lowest false discovery rate. In a similar vein, it was predicted that among the seven bioactive compounds, a (kaempferol-3-O-rutinoside) would have the biggest impact, exhibiting interactions with 11 molecules: MAPK1, NAMPT, NFKB1, PRKCD, HSP90AA1, CASP8, STAT1, TLR4 and MAPK8 based on this compound a carried out for further *in silico* evaluation studies. However, as shown in Figure 2 (A), network analysis identified eight molecules, specifically (a-h). The *Adansonia digitata* L. compound (a-h)-target network in regulating antidiabetic responses. The path sare represented by the round nodes, the targets by the down arrow form, and the molecular pathways by the diamond shape.

Gene Ontology (GO) and Kyoto Encyclopaedia of Genes and Genomes (KEGG) Pathways: An Enrichment Analysis

A total of 318 GO items, comprising 289 Biological Process (BP), 11 Cell Component (CC) and 13 Molecular Function (MF) phrases, were revealed by the GO analysis. The top ten enriched BP, CC and MF GO keywords are shown in Figure 2 (B). Negative/positive response to Stress, stimuli and chemicals were the three basic BP words. Cytoplasm, nucleus and cytosol were the three main CC terminology. The majority of the MF terms had associated with protein binding, enzyme binding, signaling receptor regulator activity, Signaling receptor binding, Identical protein binding, enzyme ion binding, phosphatase binding and nitric-oxide synthase activity. Furthermore, bioactive compounds of *Adansonia digitata* L. were found to be significantly enriched in 136 KEGG pathways. These pathways may primarily be impacted by HIF-1, C-type lectin receptor, IL-17, cAMP, apoptosis, necroptosis, digestion and absorption, among other pathways. The top 15 KEGG enrichment pathways for shared targets are displayed in Figure 2 (C). Together, these pathways aid in the body's initiation and control of DM responses.

Ramachandran plot statistics for α-amylase

A total of 495 amino acid residues were found in α-amylase (PDB: 4W93) according to the PROCHECK tool. Of these, 387 (92.1%), 33 (7.9%), 0 (0.0%) and 0 (0.0%) were distributed in the most wanted, furthermore allowed and disallowed regions, respectively. Furthermore, neither proline nor glycine was present in 420 residues. In addition, two terminal residues-glycine and proline excluded-were present. The counts for proline and glycine were 50 and 23, respectively. Figure 3 (A) showed that 3D structure and Ramachandran plot Of α-amylase (PDB:4W93). For all sorts of residues with extended side chains to possess both angles,the Chi1-Chi2 of α-amylase is Offered to take into account the Chi1-Chi2 side chain torsion angle combinations [Figure 3 (B)]. The Z-score generated was found within the range of its native proteins, which validates that the protein models used is high quality. Overall model quality (Z-score) for α-amylase (PDB:4W93) found to be -9.51 (Figure 3 C).

Molecular docking analysis

First, chemically driven a (Kaempferol-3-O-rutinoside)was docked against the chosen protein to gauge the optimal structural locations within the target proteins' active areas. Based on their bonding interaction patterns and lowest energy values (Kcal/mol), all formed docked complexes were assessed. Compound a's binding site displayed a binding affinity value of -8.21 kcal/mol and a Glide energy of -61.68 kcal/mol with eight hydrogen bond interactions between (OH) groups and residues of hydrogen

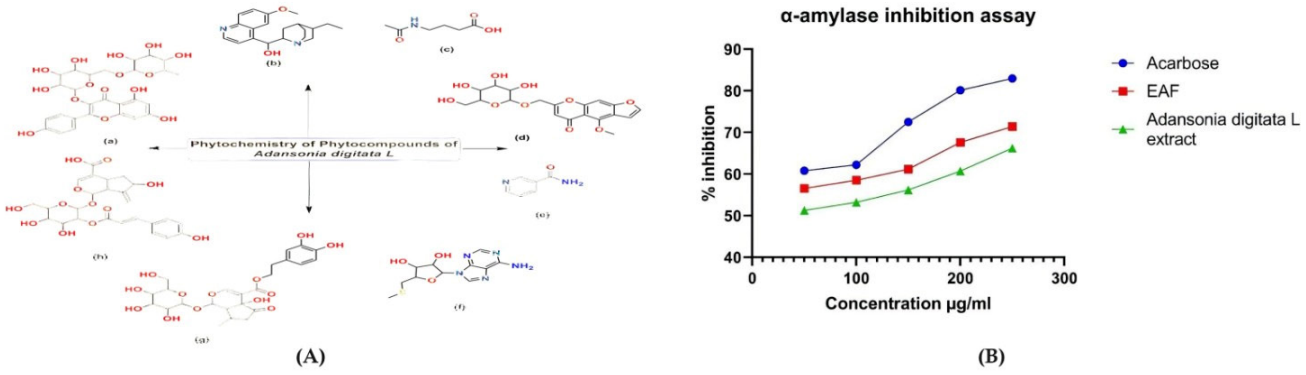

Figure 1: (A) Phytochemistry of selected Phytocompounds of *Adansonia digitata* L. (B) Determination of α-amylase inhibition assay.

bonds with GLU233, ASP197, ARG195 and Pi-Pistacking between HIP305, TRP58 and TYR62. Meanwhile, standard acarbose displayed a binding affinity value of -7.81 kcal/mol and a Glide energy of -45.06 kcal/mol with two hydrogen bond interactions between GLU240, ASP300, HIE201, ASP197, ARG195 and HIP305, respectively (Figures 4 (A) and (B), Table 2).

Prediction of Physico chemical Properties

The ADMET was predicted using Schrodinger's software's Qikprop module, which covered hydrophilicity, molecular weight, permeability, hydrogen bond donors, hydrogen bond acceptors, binding to human serum albumin and oral absorption. The anticipated and recommended values were compared with violations of Lipinski's rule of five and Jorgensen's rule of three (Table 3). The bioavailability radar graphic (Figure 4 (C)) shows that the optimal range (pink region) for each of the six main variables-lipophilicity, size, polarity, solubility and flexibility accept saturation-provides a reasonable estimate of their oral bioavailability. Contrarily, compound a remained far from the ideal regions of polarity and size and it was predicted that it would never be more orally bioavailable. Additionally, compound a's pharmacokinetic properties were analysed with regard to its potential and the functional group contributing to its non-toxic qualities is indicated by the structural colour green, while the functional group contributing to itstoxic characteristics is indicated by the structural colour brown. It was discovered that the chemical a was not toxic for acute oral toxicity.

DISCUSSION

Molecular docking has truly revolutionized diabetes research. By simulating how different compounds interact with target proteins, scientists can efficiently identify potential treatments.

Table 1: Analysis of Modified Proteins by Enrichment.

Term ID	Term description	Observed	gene count	False discovery rate	Matching proteins in network (labels)
hsa04066	HIF-1 signaling pathway.	9	102	1.03E-11	MAPK1, SERPINE, NFKB1, EP300, NOS3, NOS2, TLR4, SLC2A1, PIK3R1.
hsa04625	C-type lectin receptor signaling pathway.	7	101	9.53E-09	MAPK1, NFKB1, PRKCD, CASP8, STAT1, MAPK8, PIK3R1.
hsa04657	IL-17 signaling pathway.	5	91	4.53E-06	MAPK1, NFKB1, HSP90AA1, CASP8, MAPK8.
hsa04024	cAMPsignaling pathway.	6	207	9.35E-06	CFTR, MAPK1, NFKB1, EP300, MAPK8, PIK3R1.
hsa04210	Apoptosis	5	131	1.83E-05	MAPK1, NFKB1, CASP8, MAPK8, PIK3R1.
hsa04217	Necroptosis	5	147	3.03E-05	HSP90AA1, CASP8, STAT1, TLR4, MAPK8.
hsa04621	Digestion and absorption.	9	173	3.89E-10	MAPK1, NAMPT, NFKB1, PRKCD, HSP90AA1, CASP8, STAT1, TLR4, MAPK8.
hsa05418	Fluid shear stress and atherosclerosis.	8	129	1.50E-09	PLAT, NFKB1, NOS3, HSP90AA1, BMPR1A, MAPK8, NFE2L2, PIK3R1.
hsa04922	Glucagon signaling pathway.	4	100	0.00012	SIRT1, GCK, EP300, SLC2A1.
hsa04910	Insulin signaling pathway.	4	132	0.00032	MAPK1, GCK, MAPK8, PIK3R1.
hsa04932	Non-alcoholic fatty liver disease.	4	146	0.00046	NFKB1, CASP8, MAPK8, PIK3R1.
hsa04662	B cell receptor signaling pathway.	3	78	0.0012	MAPK1, NFKB1, PIK3R1.
hsa04750	Inflammatory mediator regulation of TRP channels.	3	92	0.0018	PRKCD, MAPK8, PIK3R1
hsa04215	Apoptosis-multiple species.	2	30	0.0041	CASP8, MAPK8.
hsa04930	Type II diabetes mellitus.	5	45	2.67E-07	MAPK1, GCK, PRKCD, MAPK8, PIK3R1.

This technique not only speeds up the drug discovery process but also provides valuable insights into the safety and effectiveness of new therapies. Recent studies highlight how bioactive compounds from traditional medicines could inhibit crucial enzymes linked to diabetes, offering promising new treatment avenues. While there are challenges-like ensuring simulated interactions mirror real biological processes-advancements in computational methods and molecular biology are paving the way for significant breakthroughs.

Herbal medicines have recently received increased attention for their different biological activities and phytometabolic contents for treating diabetes mellitus. The plant *Adansonia digitata* L. was harvested and the presence of phytocompounds was determined using hydroalcoholic extracts and previously published GC-MS analysis [Figure 1 (A)]. Secondary metabolites have a wide range of biological and medicinal properties. Phytoconstituents were measured using a variety of biochemical tests and hydroalcoholic extracts were successively liquid extracted with ethyl acetate.

Each fraction was collected, dried under low pressure and concentrated to get the Ethyl Acetate Fraction (EAF). Before being tested for α-amylase inhibitory activity against standard acarbose. The results of the experiment showed [Figure 1 (B)]. The hydroalcoholic extract and EAF showed significant inhibitory activity against αamylase, with IC_{50} values of 33.90 µg/mL and 29.21 µg/mL, correspondingly. The activities were compared to the standard acarbose, with an IC_{50} of 23.18 µg/mL for α-amylase. Further, A combined C-T and T-P network depicts the relationship between phytochemicals, protein targets and pathways. The C-T-P network had 104 edges and 21 nodes. Thirteen different pathways regulated by proteins linked to diabetes mellitus were found by gene set enrichment analysis. Peer interpretation of protein interactions using KEGG pathway analysis revealed 4 pathways that are closely related to the pathogenesis of diabetes mellitus.

Table 3: Physico chemical properties of compound a.

Sl. No.	Property	Compound a	Range
1	SASA	767.95	300 to 1000
2	dHB	8	0 to 6
3	aHB	19	2 to 20
4	QPlogPo/w	-1.8	-2.0 to 6.5
5	QPlogS	-2.2	-6.5 to 0.5
6	QPlogHERG	-5.1	Below-5
7	QPPCaco	5.26	<25 poor, >500 great
8	QPlogBB	-3.62	6ishigh,0islow
9	QPPMDCK	1.70	<25 poor, >500 great
10	QPlogKhsa	-1.197	-1.5 to 1.5
11	%Human Oral Absorption	1	>80% is high<25% is poor
12	CNS	-2	-2.0 to 2.0
13	Rule of Five	3	0 to 5
14	Rule of Three	2	<1
15	MolPSA	249.20	7-200
16	MolVol	1512.5	500 to 2000
17	DLS	0.53	-0.4to5.6

SASA: Total surface area of the solvent available in square angstroms, dHB: donor HB, aHB: acceptor HB, QPP MDCK: Apparent MDCK permeability, QPlog Khsa: Estimation of binding to albumin in human serum, QPlog BB: Anticipated partition coefficient between the brain and blood, Q Plog Po/w: Theoctanol/water partition coefficient, QPlogS: Aqueous solubility, QPlog HERG: IC_{50} value for blockage of HERG K^+channels, QPPCaco: Apparent Caco-2 permeability, HOA: Percent human oral absorption, CNS: Predicted central nervous system activity, ROF: Rule of five violations, ROT: Rule of three violations, MolPSA: Molecular polar surface area, MolVol: Molecular volume, DLS: Drug likeness score.

Table 2: The kcal/mol for compound a(Kaempferol-3-O-rutinoside)against α-amylase protein target (PDB: 4W93).

Compounds.	Glide score	Glide Energy	Docking score	No. of bonds
a (Kaempferol-3-O-rutinoside)	-8.12	-61.68	-8.21	H.b Pi 4 3
Acarbose	-7.18	-45.069	-7.81	2 0

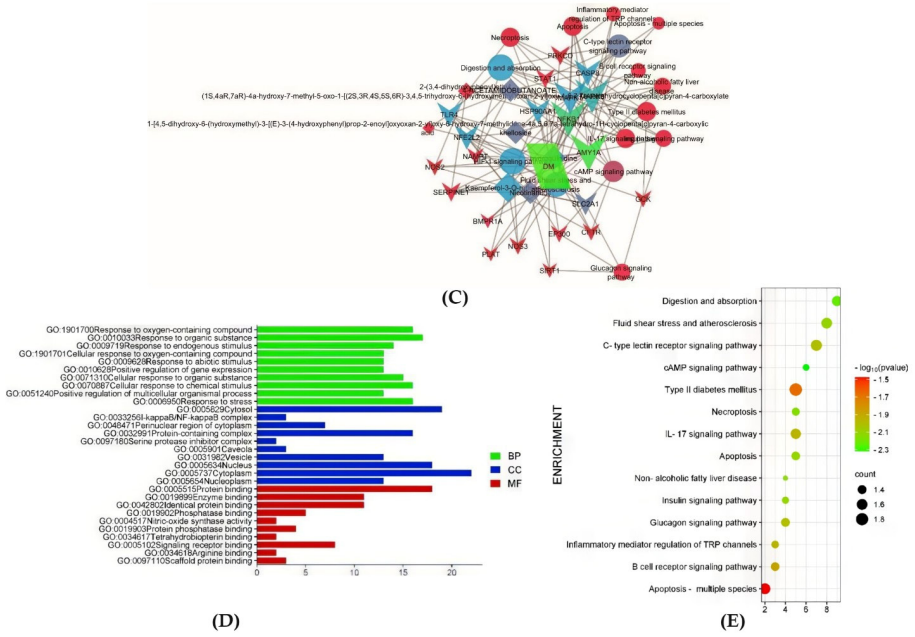

Figure 2: (A) Network representation between compound (a-h) of *Adansonia digitata* L., targets and pathways interaction. (B) BP, CP and MF gene GO analysis of proteins that are modulated. (C) The shared targets' KEGG enrichment pathway.

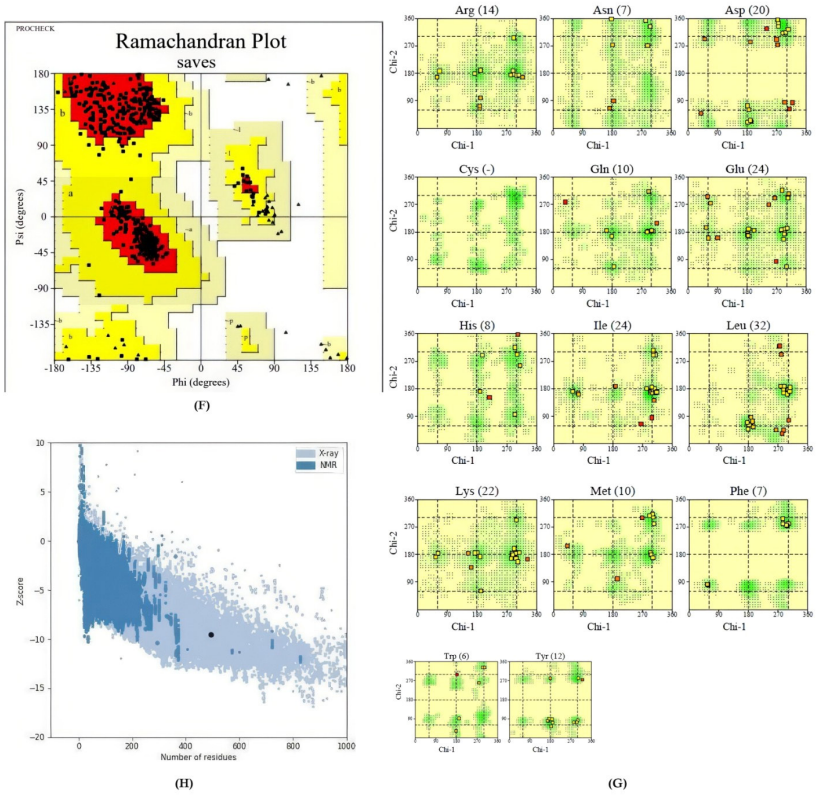

Figure 3: (A) Ramachandran plot of 4W93 receptor generated by pro check server shows 92.1% amino acids in most favored regions and 7.9% in additional allowed regions. (B) Labels are applied to those with unfavorable conformations (scoring <- 3.00).The numbers of residues for α-amylase (PDB:4W93) are shown in a Chi1-Chi2 plot with brackets. Shading reveals advantageous conformations, based on analysis of 163 structures at a resolution of 2.0Å or above. (C) Z-score plot for the 4W93 found to be -9.51 generated by ProSAweb server.

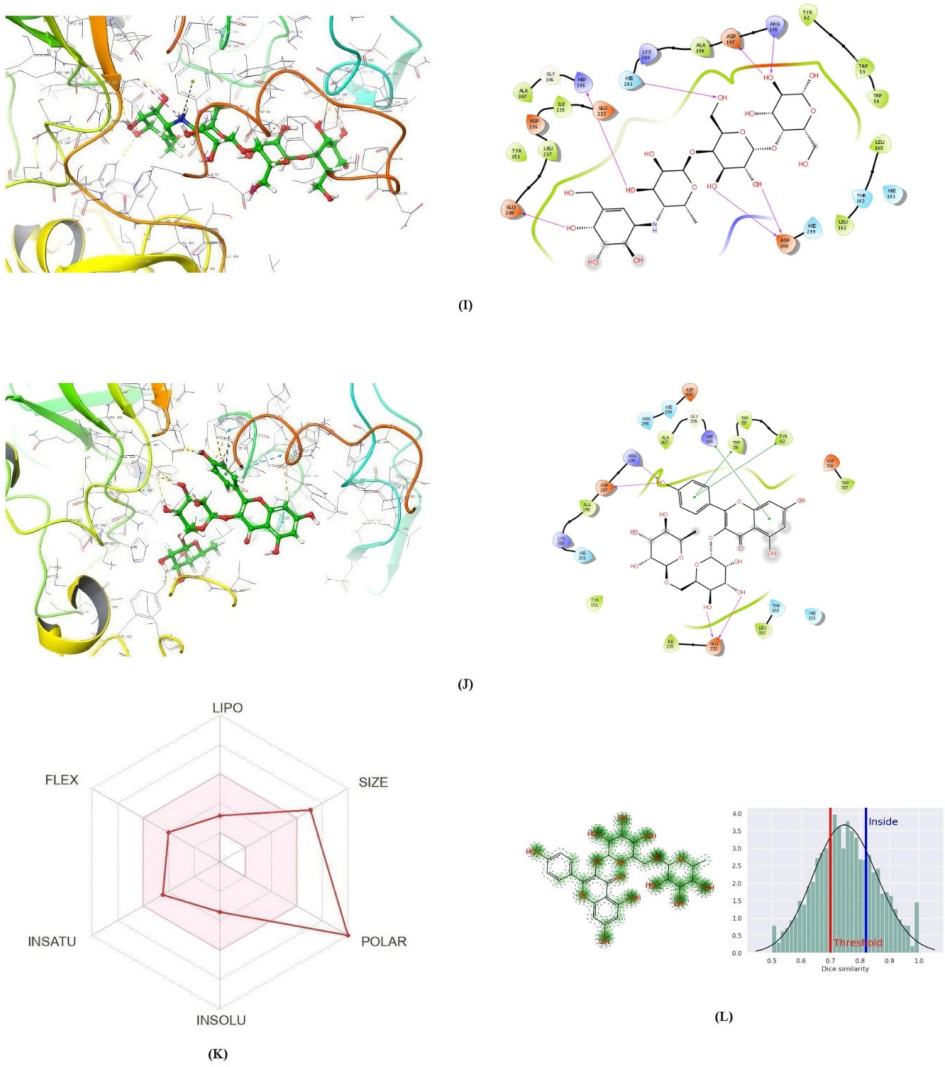

Figure 4: (A) Pose of Acarbose in α-amylase (PDB: 4W93, (B) Pose of Compound a (Kaempferol-3-O-rutinoside) in α-amylase (PDB: 4W93), (C) Bioavailability radar chart of the potent compound a. The ideal value for each oral bioavailability factor was shown in the pink region, and the expected ones for the assessed molecules were shown in red lines, (L) Acute oral toxicity of potent compound a.

Interestingly, Notably, the absorption and digestion Process had the lowest false discovery rate and the highest number of gene sets (see Table 1). Similarly, kaempferol-3-O-rutinoside was anticipated to have the most impact among the eight bioactive chemicals since it interacted with 11 distinct molecules, including MAPK1, NAMPT, NFKB1, PRKCD, HSP90AA1, CASP8, STAT1, TLR4 and MAPK8. Based on these results, compound a (kaempferol-3-O-rutinoside) was subsequently carried out for *in silico* molecular docking showed that (Table 2). To find the ideal conformation allocation inside the target protein's active domain, the chosen compounds were docked against the chosen target protein and kaempferol-3-O-rutinoside has -8.21 Kcal/mol and the oxygen atom of the molecule formed Hydrogen bonds with GLU233, ASP197 and ARG195, as well as Pi-Pi stacking of HIP305, TRP58 and TYR62 [Figure 4 (J)]. As compared to typical drugs, acarbose has -7.81 Kcal/mol and the oxygen atom of the molecule structures Hydrogen bonds with GLU240, ASP300, HIE201, ASP197, ARG195 and HIP305. [Figure 4 (I)] against the α-Amylase and the pharmacokinetic characteristics of bioactives were anticipated (Table 3).

CONCLUSION

In the present work, *in silico* gene expression, enrichment and network analysis techniques are used to examine the function of the plant *Adansonia digitata* L. as adjuvants in the management of diabetic mellitus. Additionally, the plasma protein binding model anticipated robust receptor binding, good absorption, a low blood-brain barrier, and low toxicity. These findings support the conceivable potential of compound A (kaempferol-3-O-rutinoside) as promising adjuvants in the

treatment of diabetes mellitus, as projected by the molecular docking investigations.

Since the existing results are solely dependent on database searches and knowledge-based computer simulations, they need to be more verified using with caution designed wet lab techniques, which will be the project's focus on future. This gives scientific proof to do future research, investigate the plant's lead components, examine its antidiabetic possibility using *in vivo* animal models and propose human trials.

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ABBREVIATIONS

ADMEProfile: Absorption, distribution, metabolism and excretion; **aHB:** Acceptor HB; **BP:** Biological process; **CC:** Cell component; **CNS:** Predicted central nervous system activity; **dHB:** Donor HB; **DLS:** Drug likeness score; **EAF:**Fraction of ethyl acetate; **GO:** Gene Ontology; **HOA:** Percent human oral absorption; **IC_{50}:** Half-maximal inhibitory concentration; **IL:** Interleukin; **KEGG:** Kyoto Encyclopedia of Gene and Genomes; **LC-MS:** Liquid Chromatography-Mass Spectrometry; **MF:** Molecular function; **Mol Vol:** Molecular volume; **MolPSA:** Molecular polar surface area; **OPLS33:** Optimized Potential for Liquid Simulation; **PDB:** Protein data bank; **PKa values:** Negative base-10 logarithm of the acid dissociation constant; **QPlogPo/w:** The octanol/water partition coefficient; **QPlogBB:** Anticipated partition coefficient between the brain and blood; **QPlog HERG:** IC_{50} value for blockage of HERG K^+channels; **QPlogKhsa:** Estimation of binding to albumin in human serum; **QPlogS:** Aqueous solubility; **QPP MDCK:** Apparent MDCK permeability; **QPPCaco:** Apparent Caco-2 permeability; **ROF:** Rule of five violations; **ROT:** Rule of three violations; **SASA:** Total surface area of the solvent available in square angstroms; **TPSA:** Topological Polar Surface Area; **XP:** Extra Precision.

CONFLICT OF INTEREST

The authors declare no conflict of interest. The authors have no relevant financial or non-financial interests to disclose.

SUMMARY

This study investigates the potential of *Adansonia digitata* L. in diabetes management through comprehensive phytochemical and protein analyses. Eight identified phytoconstituents, including alkaloids, terpenes and steroids, were predicted to modulate diabetic proteins. Network analysis indicated a significant role for compound A (Kaempferol-3-O-rutinoside) in interacting with key molecular targets.

Structural evaluations of α-amylase (PDB: 4W93) using PROCHECK and Chi1-Chi2 tools affirmed the protein quality. The molecular docking study revealed that compound A (Kaempferol-3-O-rutinoside) exhibited α-amylase inhibition with a binding energy of -8.21 kcal/mol, surpassing the standard drug acarbose with -7.81 kcal/mol.

ADMET predictions highlighted some deviations from ideal oral bioavailability for compound A, which underscores considerations for its pharmaceutical applications. Nonetheless, the non-toxic nature of compound A was confirmed, supporting its safety profile. The hydroalcoholic plant extract selectively inhibited α-amylase with IC_{50} values of 33.90 µg/mL. Furthermore, the Ethyl Acetate Fraction (EAF) demonstrated an IC_{50} value of 29.21 µg/mL at a concentration of 50 µg/mL, compared to the standard acarbose with an IC_{50} value of 23.18 µg/mL.

Using *in vitro* activity and computer-aided drug design models, this study unveils the promising α-amylase inhibitory characteristics of *Adansonia digitata* L. These findings provide a scientific foundation for future research, including investigating the plant's lead compounds, examining its antidiabetic potential using *in vivo* animal models and proposing human trials.

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