IN SILICO MOLECULAR PROPERTIES PREDICTION OF COMPOUNDS FROM CARICA PAPAYA LEAF EXTRACT

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ABSTRACT

Objectives: The objectives of the study were to analyze the scientific rationale and possible pharmacokinetic properties of certain biologically active compounds from *Carica papaya* leaf extract *in silico.*

Methods: This study was investigated on web-based tools PUBCHEM to extract the chemical structure followed by authentication and validation with chemical formula. The 2-dimensional structures are further converted to 3-dimensional (3D) structure with CHEMSKETCH software, the derived 3D structures are then screened for molecular properties and druglikeness score followed by absorption, distribution, metabolism, elimination, and toxicity through admetSAR software. The reports are analyzed and predicted for possible drug like compound in leaf extract of papaya.

Results: The results obtained indicate that the compounds screened papain, chymopapain, quercetin, and protocatechuic acid had the druglikeness score −0.93, −0.05, + 0.93, and +0.74, respectively. On acute toxicity prediction, quercetin belonged to Class II and other compounds belonged to Class III.

Conclusion: The study confirms the among the compounds screened for *in silico* druglikeness and adsorption, distribution, metabolism, excretion, and toxicity profile quercetin a flavonoid can be possibly developed as effective anti-dengue compound.

**Keywords:** *Carica papaya*, *In silico*, Pharmacokinetics, Toxicology.

INTRODUCTION

Carica papaya plant product extract contains many chemical constituents among which two important biologically active compounds, namely, chymopapain and papain are nowadays used for digestive disorders [1]. The papain, caricain, chymopapain, and glycine endopeptidase derived from *C. papaya* can improve gastrointestinal tract acidic pH condition and pepsin degradation. Few other active compounds of *C. papaya* considered as a "naturally immobilized" biocatalyst are lipase, a hydrolase found in water-insoluble fraction of crude papain [2]. In folklore medicine, the white papaya latex is used to treat dyspepsia and topically applied for external burns and scalds. The seeds and fruits of *C. papaya* are shade dried and powdered and used as antihelminthic as well as anti-amebic [3]. Dried and pulverized leaves decoction is administered as a purgative for horses and used for the treatment of infections of genitourinary system. The unripe and semi-ripe *C. papaya* fruits are ingested or applied on the cervix as folklore treatment to cause abortion. However, the consumption of unripe and semi-ripe papaya fruits could be unsafe during pregnancy, but consumption of ripe fruits during pregnancy causes no risk [4]. The chemical constituents individually though isolated and reported in various journals, but still individually many of these *C. papaya* compounds have not been studied for drug-like properties.

In silico computer simulation is considered as one of the most efficient ways to achieve reduction of financial burden in research, refinement, and replacement of animals in research [5]. *In silico* computer simulation of research can provide predictive results of numerous compounds of more complex phenomena for which limited or no data are available in short span of time, thereby a lead compound with high success rate can more easily be selected for further research. Hence, the current study is an attempt in search of preferred drug-like compounds from *C. papaya* leaf extract by computer simulation and an attempt to evaluate the lead compound with possible better therapeutic activity for further wet laboratory analysis. Hence, as a preliminary step, the present study is to analyze adsorption, distribution, metabolism excretion, and toxicity (ADME&T) properties of biologically active compounds from *C. papaya* leaf extract *in silico.*

MATERIALS AND METHODS

Literature Review – *Carica papaya* leaf extracts

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Selection of compounds from analysis of review article

↓

Step-1 online tool – PUBCHEM archive – Retrieval of Selected compound

↓

Download authenticated source Chemical Structure of compound

↓

Step-2 Software CHEMSKETCH-2 D to 3D Chemical structure conversion

↓

3D structure of compounds

↓

MOLsoft-software   admetSAR-software

Compound properties   Drug likeness score   ADME&T prediction

↓

RESULTS

↓

Report and Analysis of compounds

↓

In silico prediction of Drug-like candidate among screened compounds

Hardware and Software

The selected compounds molecular properties of chemical structure from leaf extract of *C. papaya* extract are carried out in Hewlett Packard 2016 Model installed with Windows 11 software, Java enabled with updated plugins.

Data Set

The selected compound chemical structures of *C. papaya* with 2-dimensional (2D) pictures were collected from accredited indexed published journal and other sources such as PubChem, Chembank, ChemPDB, and Asinex Ltd. After detailed review, the structures are drawn with ChemSketch followed by PHASE software module was used to convert the 2D structures into 3-dimensional (3D) structures [6].
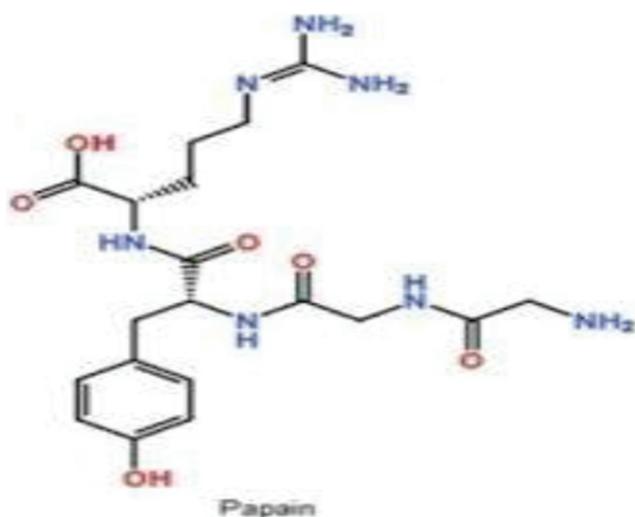
Fig. 1: Two-dimensional chemical structure of papain

Virtual screening

ADMET predictions

The 3D structures created will be used for ADMET predictions by admetSAR version 2.0. *In silico* results of compounds on absorption, distribution, metabolism, excretion, and toxicity are acquired in the virtual screening workflow protocol, followed by a web-based tool MOLSOFT L.L.C was used to run the molecular properties evaluation and druglikeness score [7,8].

Statistical methods and calculation

Calculation of molecular volume and druglikeness score is done using an interactive Molecular Properties Calculator applet (MolSoft L.L.C., San Diego, CA, USA). The study is done in the department of pharmacology and college library using online tools during July 2018–August 2018. Since the study does not involve animal and humans, exemption certificate from the Institutional Ethics Committee is obtained before initiating the study.

RESULTS AND DISCUSSION

C. papaya is an herbaceous plant known for cultivation for the production of papaya fruit. *C. papaya* leaf juice is used as an herb by Ayurvedic and Siddha systems of medicine [9]. The leaf extract preparation is used for the treatment of various ailments such as fever, inflammation, diabetes, and wound dressing [10]. Lately, Central Council for Research in Siddha has published a guideline on use of papaya leaf extract juice for Siddha practices for clinical management of dengue fever [11]. This has led to commercial production and wide use of papaya leaf extract in tablet forms as over-the-counter successful product against dengue. The plant leaf extract has many constituents and it's a challenge to identify, isolate the active principle responsible for medicinal activity. The present study by *in silico* experimental tools on human models analyzed molecular properties, ADMET, and predicted druglikeness of various constituents of papaya leaf extract which includes papain, chymopapain, quercetin, and protocatechuic acid.

The present study is designed to study and analyze the ADME&T properties followed by prediction of drug-like compound based on
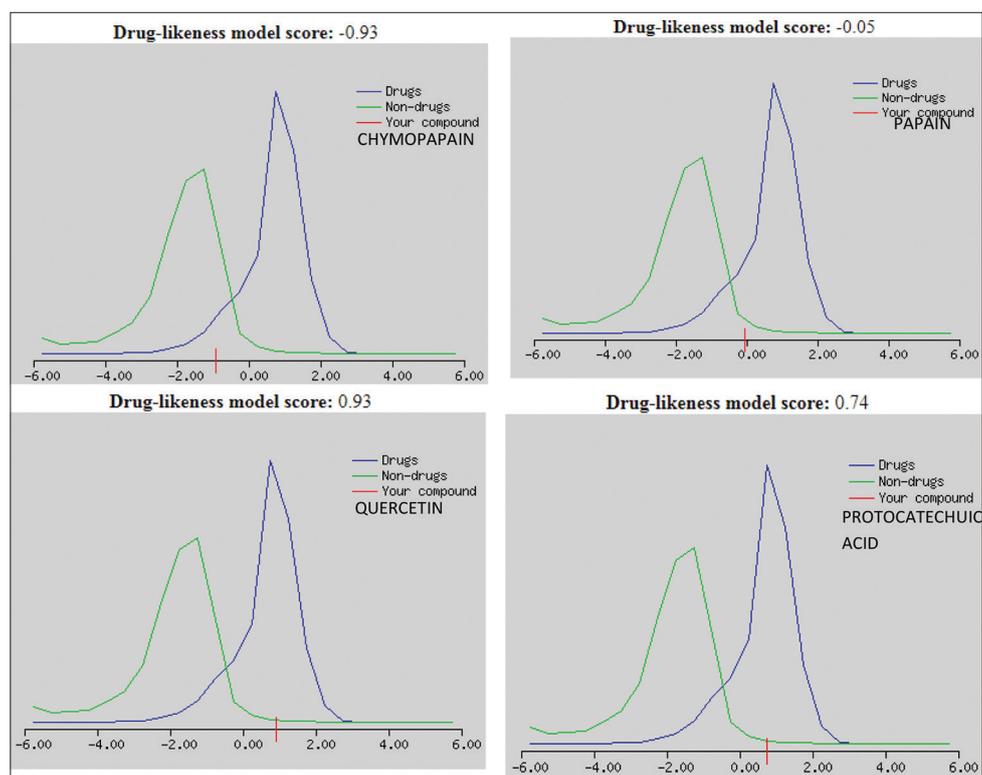

Fig. 2: Druglikeness model scoring of compounds. (a) Chymopapain, (b) papain, (c) quercetin, (d) protocatechuic acid

results obtained by *in silico* experimental models. The project was initiated after taking approval letter from the Institutional Ethics Committee on July 2018. The research was done in the department of pharmacology for a period of 25 days in the month of July 2018 to August 2018 followed by analysis and interpretation of results.

From the detailed review of literature from earlier studies, papain, chymopapain, quercetin, and protocatechuic acid 2D chemical structure were retrieved from PUBCHEM online compound database platform, all the four compounds were available with PubChem I.D for authentication and verification and successfully 2D structures converted to 3D structure by CHEMSKETCH software (Table 1), the 3D structures are the processed with MOLSOFT L.L.C software and the molecular properties are predicted (Table 2). The analysis of result for molecular properties revealed that the octanol-water partition coefficient values of quercetin (1.5) and protocatechuic acid (0.93) were

more compared with papain (−4) and chymopapain (−2) and predicted to have better water solubility of compound (Fig. 1 and 2). Among the four compounds, the hydrogen bond donor and acceptor quercetin had higher values are predicted to have better chance of getting docked to receptors as ligand (Table 2). From the analyzed report and by the structures when subjected to *in silico* druglikeness model, quercetin and protocatechuic acid had better scores +0.93 and +0.74, thereby correlating with molecular properties, quercetin was predicted as possible drug-like candidate among the screened compounds.

The ADME&T human model prediction by admetSAR software of the four compounds 3D structures was analyzed. The absorption model predicted papain and quercetin had human intestinal absorption, whereas chymopapain and protocatechuic acid had both human oral and intestinal absorption. The distribution model (Table 3) predicted all the compounds had <2% plasma protein binding (PPB), none of

Table 1: 2-dimensional structures to 3-dimensional structure conversion with ChemSketch software

S. No.	Compound	2-D structures ➡	3-D structure
1.	Papain $C_9H_{14}N_4O_3$		
2.	Chymopapain $C_6H_4Na_2O_8S_2$		
3.	Quercetin $C_{15}H_{10}O_7$		
4.	Protocatechuic acid $C_7H_6O_4$		
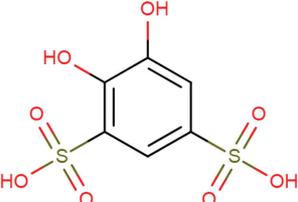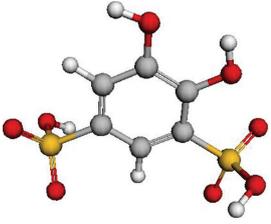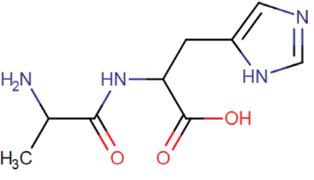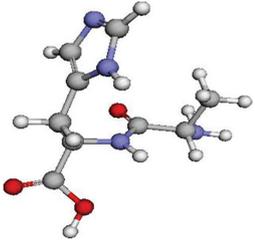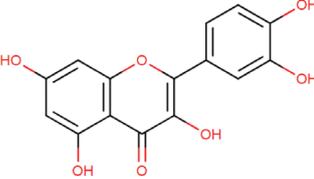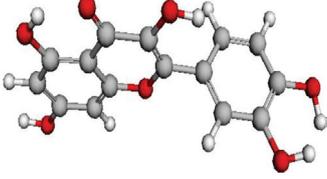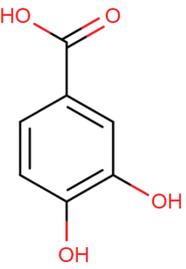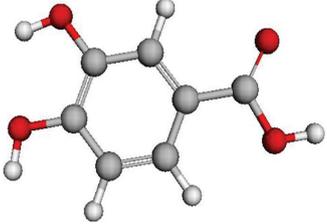
Table 2: Molecular properties and druglikeness of *C. papaya* leaf extract active compounds

S. No.	Compounds	PubChem CID	Molecular weight (g/mol)	LogP (Octanol-water partition coefficient value)	H-bond donor	H-bond acceptor	Druglikeness score
1.	Papain $C_9H_{14}N_4O_3$	9369	226.236	-4	4	5	-0.93
2.	Chymopapain $C_6H_4Na_2O_8S_2$	9001	314.19	-2	2	8	-0.05
3.	Quercetin $C_{15}H_{10}O_7$	5280343	302.238	1.5	5	7	+ 0.93
4.	Protocatechuic acid $C_7H_6O_4$	72	154.03	0.93	3	4	+0.74

CID-compound index, H-hydrogen. *C. papaya: Carica papaya*

Table 3: ADMET predicted profile of *C. papaya* leaf extract compounds

S. No.	Compounds	Absorption		
		HIA	HOB	CaCO$_2$
1.	Papain	+	−	−
2.	Chymopapain	+	+	−
3.	Quercetin	+	−	−
4.	Protocatechuic acid	+	+	

S. No.	Compounds	Distribution			
		PPB 100%	Pgp substrate	Pgp inhibitor	BBB
1.	Papain	0.581	−	−	+
2.	Chymopapain	0.744	−	−	+
3.	Quercetin	1.175	−	−	−
4.	Protocatechuic acid	0.717	−	−	−

S. No.	Compounds	Metabolism		Excretion	
		CYP 450 substrate	CYP 450 inhibitor	Plasma t1/2	Renal clearance
1.	Papain	+	−	NP	NP
2.	Chymopapain	+	+	NP	NP
3.	Quercetin	+	−	NP	NP
4.	Protocatechuic acid	+	+	NP	NP

HIA: Human intestinal absorption, HOA: Human oral bioavailability, PPB: Plasma protein binding, Pgp-P glycoprotein, BBB: Blood–brain barrier, CYP: Cytochrome, NP: Not predictable. *C. papaya: Carica papaya,* ADMET: Adsorption, distribution, metabolism, excretion, and toxicity

Table 4: Toxicology prediction profile of *C. papaya* leaf extract compounds

S. No.	Compounds	Toxicity		
		Hepatotoxicity	Ames test	Acute oral toxicity (Class)
1.	Papain	+	−	III
2.	Chymopapain	+	−	III
3.	Quercetin	+	+	II
4.	Protocatechuic acid	−	−	III

Ames mutagenicity. *C. papaya: Carica papaya*

the screened compounds are P glycoprotein substrate or inhibitor and with blood–brain barrier (BBB) model, papain and chymopapain were predicted to cross BBB. Based on poor PPB of <2% capacity of drugs, it was analyzed that all compounds have wider distribution. The metabolism model predicted all compounds are substrate of CYP450 and chymopapain and protocatechuic acid is enzyme CYP450 inhibitor. CYP 450 is one of the major enzymes responsible for metabolism and if inhibited the possibilities of drug-drug interaction side effects were predicted to be high. The elimination model has failed to predict the plasma t1/2 and renal clearance of the drug.

The toxicity model (Table 4) predicted protocatechuic acid to fall under Class III for acute oral toxicity testing and appeared safer with no hepatotoxicity and test negative for AMES. Quercetin is Class II which is positive prediction for hepatotoxicity and AMES model.

The advantage of *in silico* experimentation method is, a large number of molecules can be screened and predicted in short duration with minimal financial and human hours. The use of animals in preliminary screening can be largely reduced and by this experimental procedure no animals and humans are harmed.

Among the four compounds screened, quercetin and protocatechuic acid have preferred molecular properties and drug-like features. Quercetin a flavonoid is reported to have significant antiviral activity on dengue virus by *in vitro* testing methods [12]. There are published research articles that *C. papaya* leaf extract increased 12-lipoxygenase and platelet-activating factor receptor gene expression [13]. The *in silico* prediction data from the present study can be applied for further research on quercetin and protocatechuic acid by wet laboratory studies on above said pathways to identify platelet production

stimulation ability of the compound in treatment and management of dengue infection.

CONCLUSION

The present study selected four compounds based on extensive review of literature papain, chymopapain, quercetin, and protocatechuic acid. On the whole based on molecular properties, druglikeness score, and ADME&T model, this study concludes that quercetin and protocatechuic acid are predicted to have better drug-like features and protocatechuic acid from the papaya leaf extract had better toxicity profile.

AUTHORS' CONTRIBUTIONS

Abhigna Nara played a key role as principal investigator including drafting the proposal acquiring approval and funding from ICMR-STS Project. B.L Kudagi played a crucial role in reviewing the proposal and final approval including drafting and reviewing before and after completion of project for approval to publication. K.R.Subash contribution was toward data procurement and analysis. Sriharsha Rayam contributed as coguide for the study including drafting the article, reviewing content, final endorsement of the version to be published, and agreed to be accountable for all perspectives of the work

CONFLICTS OF INTEREST

The author declared that there are no conflicts of interest related to this study.

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