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SHORT COMMUNICATION

Insilico and Pharmacological Property Analysis of Bioactive Components from *Prunus avium* against Diabetics

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Abstract

Diabetes is a common metabolic disorder, which effects people across all cultures globally. Lifelong distress is the cause of this disease which has no cure as of now. Various medications available in the market are too expensive and not easily affordable by all. Rural people rely on plant based Ayurvedic medications to heal diabetes as these contain anti-diabetic compounds. These phytoconstituents/anthocyanin derivatives work with several mechanisms that involve phytoconstituent interactions and target molecules in diabetic metabolism. Molecular docking analysis aids in finding out the interaction between receptors and ligands to identify the finest interaction which suits the target. In this case, the study proposed examining the bonding interactions of anti-diabetic compounds/anthocyanin derivatives derived from medicinal plants (Pelargonidin, Cyanidin, Delphinidin, Peonidin, Petunidin, Quercetin and pancreatic alpha-amylase (4X9Y)) with the help of the computational tool. ADME/T test helps decide different pharmacological and physicochemical analysis of lead atoms, degree of adsorption inside the cell, digestion rate, solvency, blood cerebrum boundary penetrability, cancer-causing nature and so on, which are the significant essentials prior to advertising a medication. Peonidin and Quercetin was proposing the best interactions. Nonetheless, to discover a better cure for diabetes, further in-vitro/ in vivo studies have to be carried out.

Keywords: Anthocyanins, Diabetes, Molecular Docking, Pancreatic alpha-amylase (4X9Y), Phytochemicals, α-amylase

1. Introduction

Diabetes mellitus is a metabolic disorder such as abnormal carbohydrate metabolism, irregular food habits and other lifestyle, which are likely to affect the blood glucose level[1]. Type 2 diabetes also affects other organs like the eyes, kidney, blood vessel, heart and nerves[2]. There are different treatments available like phototherapy, enzyme inhibition, antihypertensive therapy, recombinant insulin administration etc., depending on the type of complications involved in diabetes. There are so many plants that exist here with a fair amount of healing properties for diabetes[3]. It is used for many therapeutic purposes. Anthocyanins, a group of compounds are one of the members of the flavanoid family[4,5]. It contains a large amount of polyphenol

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and phytochemicals, which are abundantly present in plant foods[6,7–10]. The foremost reason of this study is to evaluate the ethnopharmacological information of *Prunus avium*[11–13]. Online computer-based tools are used to produce safe and reduce the number of trails during drug manufacture[14–16].

1.1 Wild-type Human Pancreatic Alpha-Amylase (4X9Y)

Wild-Type Human Pancreatic Alpha-Amylase is made up of three structural domains. The X-ray diffraction has been determined to 1.8 A resolution, and the total Structure Weight is 56.01 kDa. The Domain A contains 1–99 and 169–404 residues and Domain B contains least number of residues which is form between 100-168 residue (Figure 1).

2. Materials and Methods

2.1 Protein Data Bank (PDB)

This database contains information about 3D structure of biological compounds such as proteins and nucleic acids can be found. The proteins and nucleic acids present in every organism, and understand the 3D structures and role of these compounds during drug development. The three-dimensional structure of Wild-Type Human Pancreatic Alpha-Amylase (PDB ID 4X9Y) with 496 residues is retrieved from Protein Data Bank.

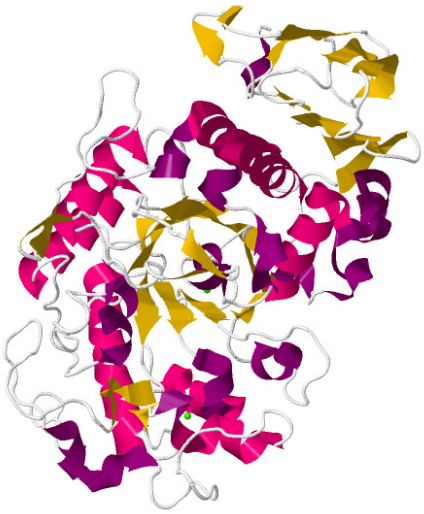


Figure 1. Three-Dimensional structure of 4X9Y.

2.2 Pubchem Database

PubChem is collecting information on chemical structures, identifiers, chemical and physical properties, biological activities, patents, health, safety, toxicity data and many others. PubChem mostly contains small molecules and larger molecules such as Chemical compounds including drugs, Nucleotides including siRNAs and miRNAs, Carbohydrates, Lipids, Peptides, Chemically-modified macromolecules.

The canonical smiles of ligand Pelargonidin (ID: 440832), Cyanidin (ID: 128861), Petunidin (ID: 441774), Peonidin (ID: 441773) and Quercetin (ID: 5280343) were collected and it is converted to pdb by using openbebel.

2.3 Active Site Identification

BiteNet (https://sites.skoltech.ru/imolecule/tools/bitenet) server is used for the identification of probable active sites.

2.4 Drug Likeness Property and ADME Prediction

SWISSADME server is used to analyze the molecular structure of ligand that confirms that the ligand using five (or) not of Lipinski's rule. At the same time, the ORISIS property explorer helps out in calculating the physicochemical properties[17,18].

Pharmacokinetics and pharmacodynamic properties were predicted using ADME/T- SAR which is an inline-based tool that include the AMES toxicity, human abdominal adsorptionetc[19].

2.5 Autodock 4.2

Autodock 4.2 is used to evaluate the performance of protein-ligand docking. The relationship between ligand and receptor was calculated by using Lamarckian algorithm (LGA). AutoGrid is used to calculate the receptor proteins represented in the grid maps.

2.6 Visualization

The docked files are visualized using Discovery Studio Visualizer, Pymol viewer, pymol and UCSF Chimera[20].

3. Results and Discussion

3.1 Active site of 4X9Y

The using BiteNet (https://sites.skoltech.ru/imolecule/tools/bitenet) the ligand-binding sites of the receptor 4X9Y was predicted (Table 1).

Lipinski's rule indicates the solubility, bioavailability and permeability of the drug compound of interest. From the analysis, Cyanidin, Pelargonidin, Quercetin, Petunidin, Peonidin follow Lipinski's rule and Lower log P indicates better absorption of the drug inside the cell. Log S value represents the drug molecule's solubility, and a lower value again reflects the candidate molecule's better solubility. Lower log P indicates better absorption of the drug molecule inside the cell. Log S value represents the drug molecule's solubility, and a lower value again reflects the candidate molecule's better solubility. TPSA or topological polar surface area is also associated with absorption and permeability of the drug molecule. Any candidate with a higher TSPA value often results in low permeability in a biological system. Quercetin and Petunidin showed the highest TPSA value (127.4 and 123.52), and Pelargonidin showed the lowest TPSA value (94.06). Peonidin and Cyanidin showed moderate levels of TPSA value - 114.29 and 103.29, respectively (Table 2).

Quercetin and Peonidin molecules are impermeable to blood-brain barriers offering no chance to cause damage to the brain. Every molecule showed Petunidin high absorption ability in the intestine and didn't show any indication of carcinogenicity. Only Petunidin and Peonidin showed colorectal tissue permeability. Cytochrome P450 (CYP) family of enzymes plays significant roles in drug metabolism, excretion and drug-drug interaction. Inhibition of any of these enzymes may cause unusual effects like low degradation rate of the drug molecule, accumulation in the body and slow excretion (Newbert and Russel, 2002). Quercetin and Peonidin are potent inhibitors of CYP450 3A4, and all molecules are inhibitors of CYP450 1A2. Gallic acid and magniferin are not any inhibitors of any cytochrome P450 enzyme (Table 3).

3.2 Docking Results

Molecular docking uses a specific algorithm to find the best ligand molecule that fits within the target molecule's active site possessing the best possible pose. The molecular docking was done to study the boding interaction between the selected ligand molecules - Cyanidin, Pelargonidin, Quercetin, Petunidin and Peonidin interacted with the target molecule with -5.5 Kcal/mol, 5.26Kcal/mol, -6.16 Kcal/mol, -2.41 Kcal/

Table 1. Active site of 4X9Y

Chain	A	A	A	A	A	A	A	A	A	A	A	A	A	A
Residues	62	162	195	197	198	233	300	318	322	385	387	388	389	390
Type	TYR	LEU	ARG	ASP	ALA	GLU	ASP	ALA	LYS	GLU	ARG	TRP	ARG	GLN

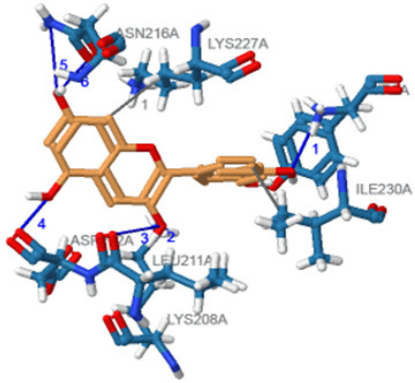
Table 2. Drug-likeness properties of selected Anthocyanin Derivatives

Drug likeness properties	Cyanidin	Pelargonidin	Quercetin	Petunidin	Peonidin
Molecular weight (g/mol)	287.24	271.24	302.2	317.27	301.27
Log P	-2.59	-2.29	1.63	-1.72	1.84
Log S	-3.34	-2.76	-3.16	-2.66	-2.81
H-bond acceptor	6	5	7	7	6
H-bond donor	5	4	5	5	4
Molar refractivity	76.17	74.15	78.03	82.66	80.64
Heavy atoms	21	20	22	23	22
TPSA	114.29 A^2	94.06A^2	127.4 A^2	123.52 A^2	103.29 A^2
Rotatable bonds	1	1	1	2	2

Table 3. ADME/T properties

Properties	Cyanidin	Pelargonidin	Quercetin	Petunidin	Peonidin
Blood-brain barrier	BBB+	BBB+	BBB-	BBB-	BBB-
Human intestinal absorption	HIA+	HIA+	HIA+	HIA-	HIA+
Caco-2 permeability	Caco2-	Caco2-	Caco2-	Caco2+	Caco2+
YP450 2C9 Substrate	Non-substrate	Non-substrate	Non-substrate	Non-substrate	Non-substrate
CYP450 2D6 Inhibitor	Non-substrate	Non-substrate	Non-substrate	Non-substrate	Non-substrate
CYP450 3A4 Inhibitor	Non-substrate	Non-substrate	Non-substrate	Non-substrate	Non-substrate
CYP450 1A2 Inhibitor	Inhibitor	Inhibitor	Inhibitor	Inhibitor	Inhibitor
CYP450 2C9 Inhibitor	Non-inhibitor	Inhibitor	Non-inhibitor	Non-inhibitor	Inhibitor
CYP450 2D6 Inhibitor	Non-inhibitor	Non-inhibitor	Non-inhibitor	Non-inhibitor	Non-inhibitor
CYP450 2C19 Inhibitor	Non-inhibitor	Inhibitor	Non-inhibitor	Non-inhibitor	Inhibitor
CYP450 3A4 Inhibitor	Non-inhibitor	Non-inhibitor	Inhibitor	Non-inhibitor	Inhibitor
CYP Inhibitory Promiscuity	High	High	High	High	High
AMES Toxicity	Non-AMES toxic	Non-AMES toxic	Non-AMES toxic	Non-AMES toxic	Non-AMES toxic
Carcinogens	Non-carcinogens	Non-carcinogens	Non-carcinogens	Non-carcinogens	Non-carcinogens
Biodegradation	Not ready	Not ready	Not ready	Not ready	Not ready
Acute Oral Toxicity	II	II	II	II	II
Carcinogenicity (Three-class)	Non-Required	Non-Required	Non-Required	Non-Required	Non-Required

mol and -6.53 Kcal/mol binding energy respectively. Cyanidin formed hydrogen bonds with 4 amino acid residues (ASN 216A (2.11), ASN 216A(3.12), ASP212A(2.17), LEU211A(3.65), LYS208A(2.09)) of the target molecule. Pelargonidin formed 5 hydrogen bonds (ASN 216A(2.06), ASN 250A(3.16), ASP212A(2.46), ILE230A(3.09), LYS208A(1.70), ARG252A(3.54)) as Gallic acid within the active site of GSK3B Quercetin formed 5 hydrogen bonds (TRP316A(2.01), LYS322A(2.04), ARG343A(1.94), ARG343A(1.99), GLN390A(2.38), GLU484A(3.12), GLU484A(3.06), GLU484(2.09)) as Gallic acid within the active site of 4X9Y. Peonidin formed 4 hydrogen bonds (GLN63A (2.25), ALA106A (3.16), ALA106A (2.13), THR163A (2.34), THR163A (1.71)) again as Gallic acid within the active site of 4X9Y. The results show that the binding energy of peonidin–4X9Y complex and Quercetin–4X9Y complex is almost equal. This could be a possible reason for resistance against the drug (Figure 2 and Table 4).



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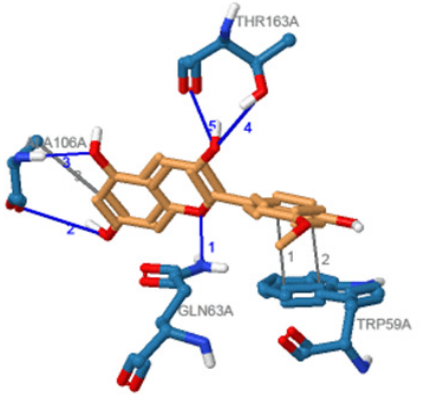

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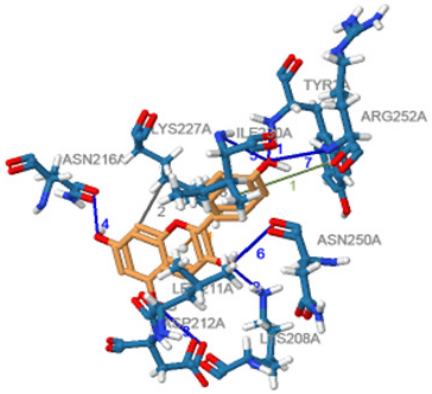

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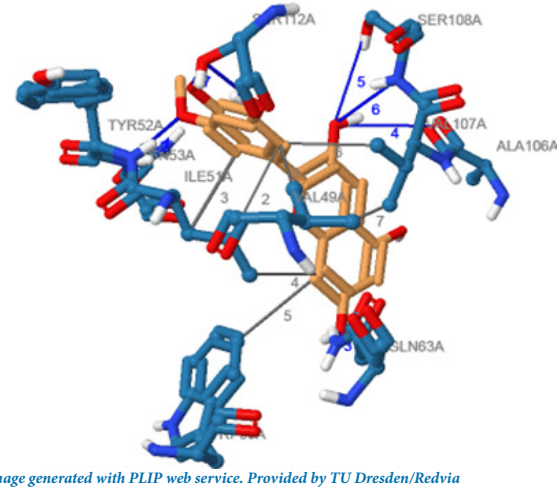

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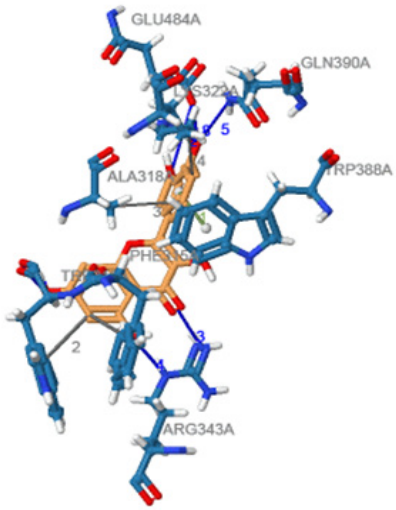

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Figure 2. 2D representation of ligand-receptor interaction: (1) Cyandin, (2) Pelargonidin, (3) Peonidin, (4) Petunidin and (5) Quercetin with 4X9Y.

Table 4. Docking results of Ligand – Receptor interaction

Compound name	ID	Docking score (Kcal/mol)	H- bonds(Å)	Bonding Residues
Cyanidin	128861	-5.7	ASN 216A(2.11), ASN 216A(3.12), ASP212A(2.17), LEU211A(3.65), LYS208A(2.09)	LYS208A, LEU211A, ASP212A, ASN 216A, ILE230A
Pelargonidin	440832	-6.9	ASN 216A(2.06),ASN 250A(3.16),ASP212A(2.46),ILE230A(3.09), LYS208A(1.70),ARG252A(3.54)	LYS208A, LEU211A, ASP212A, ASN 216, ILE230A, ASN 250A, ARG252A
Quercetin	5280343	-5.59	TRP316A(2.01),LYS322 A(2.04),ARG343A(1.94), ARG343A(1.99),GLN390 A(2.38),GLU484A(3.12), GLU484A(3.06),GLU484(2.09)	PHE315A, TRP316A, ALA318A, LYS322A, ARG343A, GLU484A and GLN390A
Petunidin	441774	-5.54	TYR52A(1.97),ASN53A(2.23),GL N63A(2.25),ALA106A(2.89),SER 108A(3.15),SER108A(2.42),SER 112A(2.11), SER112A(1.97)	VAL49A, ILE51A, ILE51A, ILE51A, TRP59A, TRP59A, VAL107A, VAL107A, TYR52A, SER108A, SER112A, ALA106A, GLN63A, ASN53A
Peonidin	441773	-6.31	GLN63A(2.25),ALA106A(3.1 6),ALA106A(2.13),THR163A (2.34),THR163A (1.71)	TRP359A, ALA106A, GLN63A, THR163A

4. Discussions

In this study, the bioactive compounds from *Prunus avium* are proficient of binding activity to 4X9Y. Thousands of plants in nature have been reported to have anti-diabetic activity. They have a different mode of action and deploy their effects by a variety of mechanisms. Selected ligands are, Quercetin and Peonidin shows better binding results to 4X9Y (except Cyanidin, Pelargonidin and Petunidin). All the selected ligand molecules in this experiment might have anti-diabetic property since all of them interacted similarly with 4X9Y. However, peonidin and Quercetin performed the best in both docking and drug-likeness property analysis and moderately well in ADME/T analysis. Pelargonidin performed well than any other ligand molecules in ADME/T analysis, but its violation of Lipinski's rule may eradicate its choice as a natural drug. So, peonidin and Quercetin could be a potent natural inhibitor of GSK3B. However, further in vivo and in vitro experiments may be required to strengthen this experiment's finding.

5. Conclusions

Quercetin and Peonidin could be the most significant source of anti-diabetic agents against type II diabetes since commercially available treatments are not cost-effective and employ some other complications. Moreover, the other three molecules also performed well, indicating their potentiality to be used as anti-diabetic agents. Well-directed research can help us find a sound and natural source of the anti-diabetic agent. Optimistically, this experiment will create research interest among researchers about natural anti-diabetic agents from medicinal plants.

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