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IN SILICO SCREENING OF PEROXISOME PROLIFERATOR ACTIVATED RECEPTOR GAMMA (PPARG)-AGONIST FROM *Eugenia jambolana* BIOACTIVE COMPOUNDS AS POTENTIAL ANTI-DIABETIC AGENT

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Abstract

Diabetes mellitus still become a serious health problem globally. The prevalence of this incidence has become increasing over the years. Current treatment has been applied, however, the new strategy is still needed to explore. Thus, in order to combat diabetes mellitus, we aimed to evaluate the potential activity of compounds from *Eugenia jambolana* stem bark as PPARG-agonist agents. *In silico* screening was performed to assess the possible and potential compounds as anti-diabetic agents. Based on our findings, we noted there are three potential compounds as PPARG-agonist, namely ellagic acid, myricetin, and friedelin. These compounds were selected based on the binding affinity scores and interaction patterns against the target protein. In detail, the ligand-protein interaction of these three compounds are ellagic acid - PPARG interaction (Van der Waals, Conventional Hydrogen Bond, Pi-Anion, and Pi-Alkyl); myricetin - PPARG interaction (Van der Waals, Conventional Hydrogen Bond, Pi-Sulfur, and Pi-Alkyl); and friedelin - PPARG interaction (Van der Waals and Alkyl/ Pi-Alkyl). From this finding, extensive research is needed to evaluate the biological function of these ligands as an anti-diabetic agent.

INTRODUCTION

The number of diabetes mellitus incidence is increasing over the years. The World Health Organization predicts the number of diabetes reach to 300 million in 2025 [1]. As one of the biggest health problems, many scientists or clinicians have been working to suppress the incidence of diabetes mellitus. Several modalities have been proposed against this entity, however, the effective ones are still needed to explore. The peroxisome proliferator-activated receptors (PPARs) protein family consists of three different types, namely PPAR alfa (PPARA), PPAR gamma (PPARG), and PPAR delta (PPARD). PPARs have a pivotal role in several biological processes such as pattern formation, metabolic homeostasis, and cellular differentiation [2]. Current treatment of diabetes mellitus showed the activation of PPARG can lower hyperglycemia, increase insulin sensitivity, and decrease plasma triacylglycerides and free fatty acids. However, the current use of PPARG agonist is under review due to its toxic and other adverse effects [3,4].

Herbs are an alternative medicine among all the modern synthetic drugs [5]. Despite its traditional uses as food sources, herbs have numerous bioactive compounds and pharmaceutical advantages against multiple kinds of diseases [6]. *Eugenia jambolana* is a cosmopolitan plant that belongs to the Myrtaceae family. *Eugenia jambolana* is widely found in the Asia region. Almost all parts of this tree such as barks, leaves, and seeds have an anti-inflammatory

effect [7]. Other studies also stated that the barks, leaves, and seeds of *Eugenia jambolana* have a wide spectrum of medical advantages such as anti-diabetic, antibacterial, and anti-human immunodeficiency [8,9]. More specifically, the *Eugenia jambolana* stem bark contains numerous bioactive compounds such as friedelin, friedelan-3-α-ol, betulinic acid, β-sitosterol, kaempferol, β-sitosterol-d-glucoside, gallic acid, ellagic acid, gallotannin, ellagitannin, and myricetin [10]. Thus, in order to combat diabetes mellitus, we aimed to evaluate the potential activity of compounds from *Eugenia jambolana* stem bark as PPARG-agonist agents.

MATERIALS AND METHODS

The stem bark of *Eugenia jambolana* has been reported to have numerous bioactive compounds [10]. In this study we evaluate the *Eugenia jambolana* bioactive compounds from stem bark such as friedelin, friedelan-3-α-ol, betulinic acid, β-sitosterol, kaempferol, β-sitosterol-d-glucoside, gallic acid, ellagic acid, gallotannin, ellagitannin, and myricetin as mentioned in the above explanation (Table 1). The 2D structure of each ligand was retrieved from PubChem Database (https://pubchem.ncbi.nlm.nih.gov/). Then each of these compounds was evaluated based on the Lipinski role of five (http://www.scfbio-iitd.res.in/software/drugdesign/lipinski.jsp) [11]. After that, 3D protein structure was designed through SWISS-Model Webserver (https://swissmodel.expasy.org/). Molecular docking was applied to predict the binding affinity and ligands-protein interaction. All procedures in this study were based on our previous protocols [12-14].

Table 1. List of *Eugenia jambolana* stem bark bioactive compounds followed with its physicochemical properties

No.	Bioactive Compounds	CID	Molecular Mass (Dalton)	Hydrogen Bond Donor	Hydrogen Bond Acceptors	High Lipophilicity (LogP)	Molar Refractivity
1.	Friedelin	91472	426	0	1	7.98	153.35
2.	Friedelan-3-α-ol	348029	428	1	1	8.04	155.71
3.	Betulinic acid	64971	456	2	3	7.18	152.78
4.	β-sitosterol	222284	312	5	6	-0.05	77.14
5.	Kaempferol	5280863	286	4	6	0.64	62.82
6.	β-sitosterol-D-glucoside	12309060	312	5	6	-0.05	77.14
7.	Gallic acid	370	170	4	5	-0.18	32.57
8.	Ellagic acid	5281855	302	4	8	0.20	58.00
9.	Gallotannin	16133691	312	5	6	-0.05	77.14
10.	Ellagitannin	101601927	312	5	6	-0.05	77.14
11.	Myricetin	5281672	318	6	8	0.22	65.24

RESULTS AND DISCUSSION

The increasing trends of diabetes mellitus have emerged. This condition leads many researchers to find new anti-diabetic agents, one of them by optimizing the use of medicinal plants [15]. In this present study, we observed the possible and potential bioactive compounds of *Eugenia jambolana* as PPARG agonists based on their binding affinity scores and ligand-protein interaction. Numerous reports have shown that the extract from *Eugenia jambolana* has multiple advantages, especially as an anti-diabetic treatment [10,16]. PPARG is one of the nuclear hormone receptors which has full function in biological processes such as inflammation, cell growth, adipocyte differentiation, and insulin sensitivity [17]. PPARG-agonist like thiazolidinedione is used as a current treatment in diabetes

mellitus due to its application in lowering blood glucose level and reducing insulin resistance [18,19]. However, this existing application of PPARG agonist is under review due to its adverse effects [3,4]. Therefore, finding new potential PPARG agonists that are effective and safer is necessary needed.

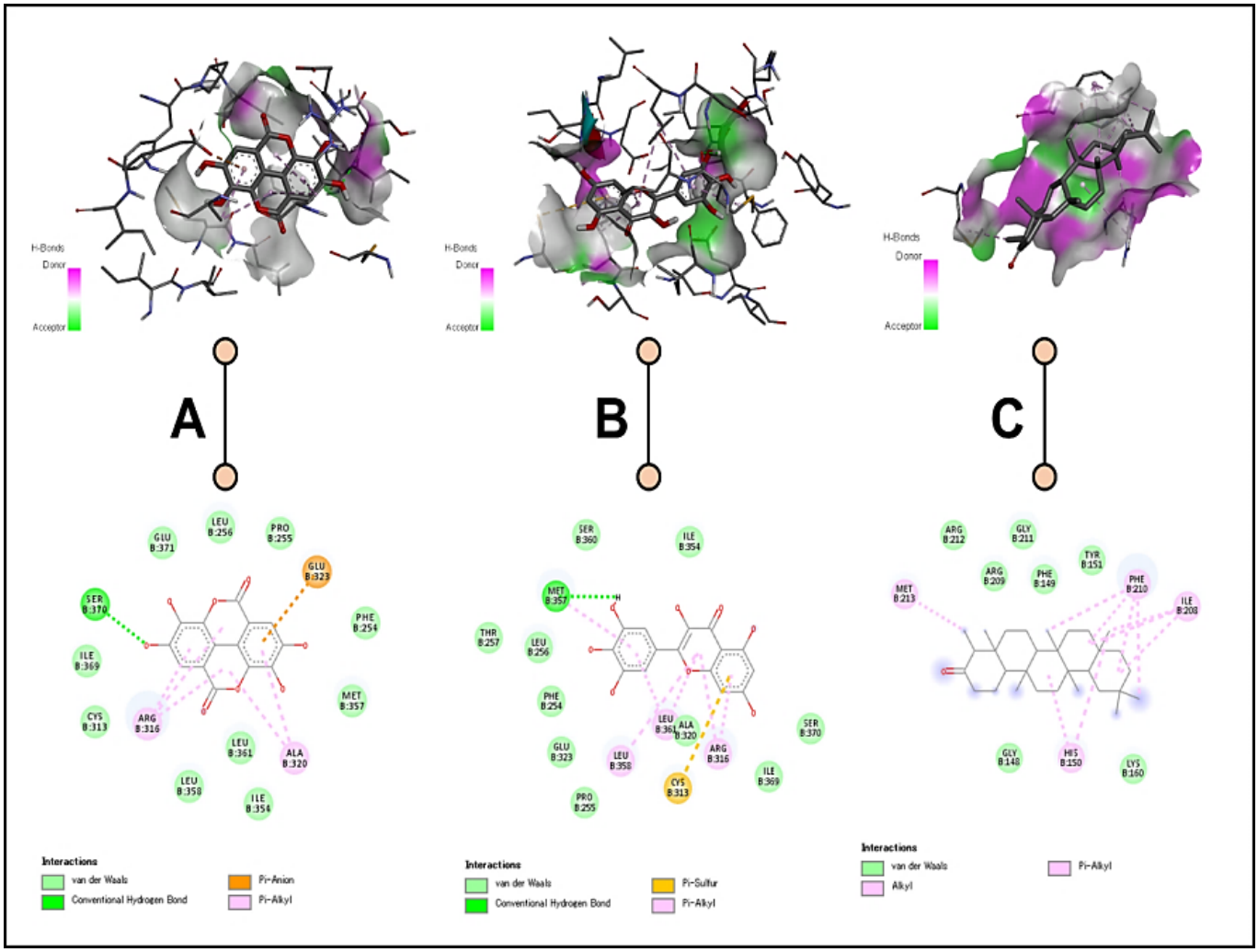

Figure 1. Visualization of predicted interaction among ligands from *Eugenia jambolana* stem bark to a target protein. A). Ellagic acid - PPARG interaction; B). Myricetin - PPARG interaction; and C). Friedelin - PPARG interaction.

In silico screening based on molecular docking among ligands and target protein demonstrated that three compounds of *Eugenia jambolana* might have great potential as an anti-diabetic (Figure 1). The three compounds include ellagic acid, myricetin, and friedelin had the most significant binding affinity score -8.2, -8.0, -7.9 kcal/mol, respectively. The lower the binding affinity score of ligands, the most favorable they interact to target protein [20,21]. Ellagic acid has been known as an antioxidant compound, which is primarily found in numerous fruits including berries, grapes, walnuts, and pomegranates. Major investigations showed that the ellagic acid possesses anti-inflammatory and antiproliferative effects. A recent study conducted by Kang et al., (2016) showed the potency of ellagic acid as anti-obesity by lessening the adiposity by the estimated doses around 30-850 mg of ellagic acid/day in an average 70-kg individual [22]. On the other hand, we also demonstrated the predicted interaction among the ligands and residual amino acid of target protein, the PPARG. Each ligands has specific interaction such as ellagic acid- PPARG interaction (Van der Waals, Conventional Hydrogen Bond, Pi-Anion, and Pi-Alkyl), myricetin - PPARG interaction (Van der Waals, Conventional Hydrogen Bond, Pi-Sulfur, and Pi-Alkyl), and friedelin - PPARG interaction (Van der Waals and Alkyl/ Pi-Alkyl) (Table 2). The chemical bond from the ligand-protein interaction is essential to keep the interaction stable.

Table 2. Binding affinity scores and the interaction prediction of potential compounds with the target protein, PPARG

No.	Ligands	Binding Affinity	Ligand - Amino Acid Interaction
1.	Ellagic acid	-8.2 kcal/mol	**Van der Waals:** GLU B:371, LEU B:256, PRO B:255, PRO B:254, MET B:357, LEU B:361, ILE B: 354, LEU B:358, CYS B:313, ILE B:369 **Conventional Hydrogen Bond:** SER B:370 **Pi-Anion:** GLU B:323 **Pi-Alkyl:** ARG B:316, ALA B:320
2.	Myricetin	-8.0 kcal/mol	**Van der Waals:** SER B:360, ILE B:354, SER B:370, ILE B:369, ALA B:320, PRO B:255, GLU B:323, PHE B:254, LEU B:256, THR B:257 **Conventional Hydrogen Bond:** MET B:357 **Pi-Sulfur:** CYS B:313 **Pi-Alkyl:** ARG B:316, LEU B:361, LEU B:358
3.	Friedelin	-7.9 kcal/mol	**Van der Waals:** ARG B:212, ARG B:209, GLY B:211, PHE B:149, TYR B:151, LYS B:160, GLY B:148 **Alkyl/ Pi-Alkyl:** PHE B:210, ILE B:208, HIS B:150, MET B:213

CONCLUSION

According to our study, we found there are three potential compounds as PPARG-agonist, namely ellagic acid, myricetin, and friedelin. These compounds were selected based on the binding affinity scores and interaction patterns against the target protein. In detail, the ligand-protein interaction of these three compounds are ellagic acid - PPARG interaction (Van der Waals, Conventional Hydrogen Bond, Pi-Anion, and Pi-Alkyl); myricetin - PPARG interaction (Van der Waals, Conventional Hydrogen Bond, Pi-Sulfur, and Pi-Alkyl); and friedelin - PPARG interaction (Van der Waals and Alkyl/ Pi-Alkyl). From this finding, extensive research is needed to evaluate the biological function of these ligands as an anti-diabetic agent.

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CONFLICT OF INTEREST

We declare there is no conflict of interest in this study.

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