COMPUTATIONAL STUDIES OF SOME POTENTIAL NATURAL PRODUCT AGONISTS ON HUMAN PEROXISOME PROLIFERATOR-ACTIVATED RECEPTOR GAMMA (PPAR-γ)

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ABSTRACT

Diabetes mellitus (DM) and diabetes insipidus (DI) are two distinct clinical entities with overlapping symptoms of polyuria and polydipsia, but diverging pathophysiology. While DI is primarily linked to either a deficiency or renal insensitivity to antidiuretic hormone (ADH), DM results from impaired insulin secretion or action. Among the regulators of metabolic homeostasis, peroxisome proliferator-activated receptors (PPARs)—particularly PPAR-γ—play a pivotal role in lipid and glucose metabolism and have emerged as key therapeutic targets for type 2 diabetes. In this study, molecular docking of ten selected natural compounds with human PPAR-γ receptor (PDB ID: 4PRG) was conducted using AutoDock VINA to evaluate their potential agonistic activity. The compounds were chosen based on reported antidiabetic activity and sourced from PubChem. The binding pocket was defined using the co-crystalized ligand 072, and interactions within the active site were analyzed. Results suggest several natural compounds demonstrate promising binding affinities and structural compatibility with PPAR-γ, offering scope for further investigation as selective PPAR-γ modulators (SPPARMs).

Keywords

Diabetes mellitus, Diabetes insipidus, PPAR-γ agonists, Molecular docking, Natural compounds, Peroxisome proliferator-activated receptors, Selective PPAR modulators (SPPARMs), Type 2 diabetes

Introduction

Diabetes Insipidus (DI)

Diabetes Insipidus is a disorder marked by extreme thirst and the frequent passage of large amounts of diluted urine. Notably, reducing fluid intake does not improve the condition. DI can occur for various reasons, and its types differ based on the underlying cause. The most common form is central (or neurogenic) DI, which results from inadequate secretion of arginine vasopressin (AVP), also called antidiuretic hormone (ADH). Another common form is nephrogenic DI, where the kidneys fail to respond properly to ADH. This type may sometimes result from the side effects of certain medications (iatrogenic cause).

The occurrence of DI is relatively rare, affecting approximately 3 in every 100,000 people. The term "diabetes" originates from the Greek word meaning "siphon," referring to excessive urination, while "insipidus" means "tasteless," highlighting the absence of glucose in the urine. DI is mainly caused by either a problem in the production of ADH or a defect in the kidney’s ability to respond to it.

Types of Diabetes Insipidus:

Central (Neurogenic) DI

This type results from damage to the hypothalamus or pituitary gland, leading to insufficient AVP production. Since AVP helps the kidneys retain water and concentrate urine, its deficiency results in increased urine output and dehydration.

Dipsogenic DI

This form is due to a malfunction in the thirst regulation center of the hypothalamus, causing an abnormal urge to drink water. Excessive water intake suppresses AVP release, leading to polyuria. Treatment with desmopressin is ineffective and may lead to water

intoxication due to continued excessive fluid intake.

Nephrogenic DI

Here, the kidneys become resistant to the effects of ADH, despite its adequate or even elevated levels in the body. This resistance impairs water reabsorption and leads to dilute urine output. It is often a result of genetic factors or drug-induced renal dysfunction.

Diabetes Mellitus (DM)

Diabetes mellitus refers to a group of chronic metabolic disorders characterized by elevated blood glucose levels, either due to insufficient insulin production or ineffective insulin utilization. Common symptoms include frequent urination (polyuria), excessive thirst (polydipsia), and increased hunger (polyphagia).

1. Type 1 Diabetes Mellitus (T1DM)

Also known as Insulin-Dependent Diabetes Mellitus (IDDM), Type 1 diabetes arises from autoimmune destruction of insulin-producing beta cells in the pancreas. This results in complete insulin deficiency. It is mostly seen in children and young adults and accounts for around 10% of all diabetes cases in Western countries. Affected individuals are often of normal weight and generally in good health at onset. There is no known way to prevent Type 1 diabetes.

2. Type 2 Diabetes Mellitus (T2DM)

This is the most prevalent form of diabetes, accounting for about 90% of cases. In Type 2 diabetes, insulin levels may remain high, but the body becomes resistant to insulin's effects. Commonly referred to as non-insulin-dependent diabetes or adult-onset diabetes, it typically develops gradually in middle-aged and older adults, often in those who are overweight. However, increasing numbers of young individuals are now also affected. Key risk factors include family history, obesity, sedentary lifestyle, and poor nutrition. Recent research also links intrauterine growth restriction (IUGR) and prenatal nutrient deficiencies to a higher risk of developing T2DM later in life.

3. Gestational Diabetes Mellitus (GDM)

Gestational diabetes occurs during pregnancy, usually after the 24th week, and shares similarities with Type 2 diabetes, involving reduced insulin sensitivity and inadequate insulin secretion. It affects around 2–5% of pregnancies and typically resolves after childbirth. However, it significantly increases the mother's risk of developing Type 2 diabetes later in life. Gestational diabetes may be triggered by hormonal changes and weight gain during pregnancy. In some cases, the placenta produces an enzyme called vasopressinase, which breaks down ADH, potentially worsening fluid regulation. Babies born to mothers with uncontrolled gestational diabetes are at risk for macrosomia (large birth weight), organ immaturity, and other complications.

The peroxisome proliferator-activated receptors (PPARs)

Peroxisome proliferator-activated receptors are nuclear hormone receptors of the NR1C family, with diverse roles regulating lipid homeostasis, cellular differentiation, proliferation and the immune response. PPARs have many potential endogenous agonists, prostacyclin, many fatty acids and their oxidation products, lysophosphatidic acid and leukotriene B4. Bezafibrate acts as a non-selective agonist for the PPAR family. These receptors also bind hypolipidemic drugs (PPARα) and anti-diabetic thiazolidinediones (PPARγ), as well as many non-steroidal anti-inflammatory drugs, such as sulindac and indomethacin. Once activated by a ligand, the receptor forms a heterodimer with members of the retinoid X receptor family and can act as a transcription factor. Although radioligand binding assays have been described for all three receptors, the radioligands are not commercially available. Commonly, receptor occupancy studies are conducted using fluorescent ligands and truncated

forms of the receptor limited to the ligand binding domain.

Peroxisomes are subcellular organelles found in most plant and animal cells that perform diverse metabolic functions including H2O2-based respiration, β-oxidation of fatty acids (FAs), and cholesterol metabolism. Peroxisome proliferator-activated receptors (PPARs) proteins belong to superfamily of phylogenetically related protein termed nuclear hormone factor. PPARs were identified in rodents in 1990 and these belong to a nuclear hormone receptor superfamily containing 48 members. But these agents are associated with no proliferation in the human beings. Structurally, PPARs are similar to steroid or thyroid hormone receptor and are stimulated in response to small lipophilic ligands. In rodents, a large class of structurally related chemicals including herbicides, industrial solvents, and hypolipidemic drugs lead to significant increase in the number and size of peroxisomes in the liver and may cause liver hypertrophy, liver hyperplasia, hepato carcinogenesis, and transcription of genes encoding proximal enzymes. PPARs mainly exist in three subtypes; α, β/δ, and γ, each of which mediates the physiological actions of a large variety of FAs and FA-derived molecules. Activated PPARs are also capable of transcriptional repression through DNA-independent protein-protein interactions with other transcription factors such as NFκB signal activators and transducers of transcription STAT-1 and AP-1 signaling.

Structure

The PPARs possess the canonical domain structure common to other nuclear receptor family members, including the amino-terminal AF-1 trans activation domain, followed by a DNA-binding domain, and a dimerization and ligand-binding domain with a ligand-dependent trans activation function AF-2 located at the carboxy-terminal region.

PPAR isoforms

PPARs are transcription factors that belong to the Superfamily of nuclear receptors. Other members of this family include retinoic acid, estrogen, thyroid, vitamin D, and glucocorticoid receptors, and several other proteins involved in xenobiotic metabolism PPARs act on DNA response elements as heterodimers with the retinoid X receptor (RXR). Their natural activating ligands are lipid-derived substrates. The family of PPARs is represented by the following three members: PPAR-α, PPAR-δ, and PPAR-γ. They play an essential role in energy metabolism; however, they differ in the spectrum of their activity–PPAR-γ regulates energy storage, whereas PPAR-α is expressed predominantly in the liver, and to a lesser extent, in muscle, in the heart, and in bone and PPAR-δ present ubiquitously expressed in whole body regulate energy expenditure; expression of PPAR-γ in endothelial cells, vascular smooth muscle cells. PPAR-γ is further subdivided in four isoforms.

γ1 - expressed in virtually all tissues, including heart, muscle, colon, kidney, pancreas, and spleen

γ2 - expressed mainly in adipose tissue (30 amino acids longer)

γ3 - expressed in macrophages, large intestine, and white adipose tissue

γ4 - expressed in endothelial cells

Mechanism of action

PPARs function as heterodimer in association with co- activator complex that binds to DNA sequence termed peroxisome proliferators response elements (PPREs) present in promoter of target genes which leads to transactivation and trans repression of various genes. In the absence of the ligands, these heterodimers are associated with co- repressor complex which block gene transcription. Some of the agonists of various PPARs receptors are given in Balakumar P. Like PPARs, RXR exists as three distinct isoforms: RXR-α, β, and γ, all of which are activated

by the endogenous agonist 9-cis retinoic acid. No specific roles have yet been elaborated. However, synthetic RXR agonists can activate the complex and thereby obtain antidiabetic outcomes similar to those seen with PPAR agonists in mouse models of type 2 diabetes. The LBD facilitates the heterodimerization of PPARs with RXR and the resultant heterodimer subsequently binds to PPRE with the recruitment of cofactors.

Natural Compounds as potential agonists of human PPAR-gamma

Medicinal plants have been used to treat various diseases for thousands of years, and since the 19th century many bio-active pure compounds isolated from these plants became very successful drugs. Moreover, still today natural products are an important source for the discovery and development of new drugs. Natural products possess a high chemical scaffold diversity and are evolutionary optimized to serve different biological functions, conferring them a high drug-likeness and making them an excellent source for identification of new drug leads. The traditional use of plant preparations can often give strong hints for the pharmacological effects of their ingredients. A study examining 119 clinically used plant-derived drugs found that 74% of them were indeed used for disease indications related to the traditional use of the medicinal plants from which the substances were isolated. Not surprisingly, significant research efforts were undertaken to explore the PPARg activating potential of a wide range of natural products originating from medicinal plants.

Natural products prove to be a rich source for the discovery of novel PPARg ligands and many structurally diverse agonists of this receptor were recently identified from traditionally used medicinal plants or food sources. Interestingly, the majority of identified natural compounds are rather weak agonists of PPARg , often activating the receptor as partial agonists, with activation pattern distinct from the full thiazolidinediones agonists and more similar to endogenous ligands with weaker activation potential such as fatty acids and prostanoids. Noteworthy, several PPARg agonists were identified in plants used as culinary spices, beverages or food sources, opening the possibility to consider modulation of the activity of this nuclear receptor through dietary interventions. While most of the identified natural products only activate PPARg as SPPARMs, some are dual agonists able to also activate PPARa.

Aim and Objectives

Computational studies of some potential natural product agonists on human peroxisome proliferator-activated receptor gamma (PPAR-γ)

Selection of human PPAR-γ receptor crystal structures from PDB

Structural analysis of the crystal structures and their binding site characterization

Selection of some potential natural compound agonists reported in literature

Molecular docking studies of these agonists in the active site of PPAR-γ

Post docking analysis and prediction of natural compounds as better agonists

Review of Literature

The NCBI literature database PubMed was searched for finding the literature featuring research citing the relatedness of PPAR and diabetes mellitus. A few literature featuring their story and the use of different natural compounds and other agonists are given below.

The inflammatory process associated with obesity mainly arises from white adipose tissue (WAT) alterations. In the last few years, nutritional-based strategies have been positioned as promising alternatives to pharmacological approaches against these pathologies. Our aim was to determine the potential of a rice bran enzymatic extract (RBEE)-supplemented diet in the prevention of metabolic,

biochemical and functional adipose tissue and macrophage changes associated with a diet-induced obesity (DIO) in mice. C57BL/6J mice were fed high-fat diet (HF), 1 and 5 % RBEE-supplemented high-fat diet (HF1 % and HF5 %, respectively) and standard diet as control. Serum cardiometabolic parameters, adipocytes size and mRNA expression of pro-inflammatory biomarkers and macrophage polarization-related genes from WAT and liver were evaluated. RBEE administration significantly decreased insulin resistance in obese mice. Serum triglycerides, total cholesterol, glucose, insulin, adiponectin and nitrites from treated mice were partially restored, mainly by 1 % RBEE-enriched diet. The incremented adipocytes size observed in HF group was reduced by RBEE treatment, being 1 % more effective than 5 % RBEE. Pro-inflammatory biomarkers in WAT such as IL-6 and IL-1β were significantly decreased in RBEE-treated mice. Adiponectin, PPARγ, TNF-α, Emr1 or M1/M2 levels were significantly restored in WAT from HF1 % compared to HF mice. RBEE-supplemented diet attenuated insulin resistance, dyslipidemia and morphological and functional alterations of adipose tissue in DIO mice. These benefits were accompanied by a modulating effect in adipocytes secretion and some biomarkers associated with macrophage polarization. Therefore, RBEE may be considered an alternative nutritional complement over metabolic syndrome and its complications.

Recent studies show that brown rice improves glucose intolerance and potentially the risk of diabetes, although the underlying molecular mechanisms remain unclear. One of the phytochemicals found in high concentration in brown rice is γ-oryzanol (Orz), a group of ferulic acid esters of phytosterols and triterpene alcohols. Here, we found that Orz stimulated differentiation of 3T3-L1 preadipocytes and increased the protein expression of adipogenic marker genes such as peroxisome proliferator-activated receptor gamma (PPAR-γ) and CCAAT/enhanced binding protein alpha (C/EBPα). Moreover, Orz significantly increased the glucose uptake in insulin-resistant cells and translocation of glucose transporter type 4 (GLUT4) from the cytosol to the cell surface. To investigate the mechanism by which Orz stimulated cell differentiation, we examined its effects on cellular signaling of the mammalian target of rapamycin complex 1 (mTORC1), a central mediator of cellular growth and proliferation. The Orz treatment increased mTORC1 kinase activity based on phosphorylation of 70-kDa ribosomal S6 kinase 1 (S6K1). The effect of Orz on adipocyte differentiation was dependent on mTORC1 activity because rapamycin blocks cell differentiation in Orz-treated cells. Collectively, our results indicate that Orz stimulates adipocyte differentiation, enhances glucose uptake, and may be associated with cellular signaling mediated by PPAR-γ and mTORC1.

Dysregulated metabolism is implicated in obesity and other disease conditions like type 2 diabetes mellitus and cardiovascular diseases, which are linked to abnormalities of peroxisome proliferator-activated receptor gamma (PPARγ). PPARγ has been the focus of much research aimed at managing these diseases. Also, germinated brown rice (GBR) is known to possess antidiabetic, antiobesity and hypocholesterolemic effects. We hypothesized that GBR bioactive compounds may mediate some of the improvements in metabolic indices through PPARγ modulation. Cultured HEP-G2 cells were treated with 50 ppm and 100 ppm of extracts from GBR (GABA, ASG and oryzanol) after determination of cell viabilities using MTT assays. Results showed that all extracts upregulated the expression of the PPARγ. However, combination of all three extracts showed downregulation of the gene, suggesting that, in combination, the effects of these bioactives differ from their individual effects likely mediated through competitive inhibition of the gene. Upregulation of the gene may have therapeutic potential in diabetes mellitus and cardiovascular

diseases, while its downregulation likely contributes to GBR's antiobesity effects. These potentials are worth studying further.

Rice bran contains important bioactive phytochemicals. Among these phytochemicals, steryl ferulates including γ-oryzanol and its major components such as cycloartenyl ferulate (CAF), 24-methylenecycloartanyl ferulate (24-mCAF), β-sitosteryl ferulate (β-SF), and campesteryl ferulate have been intensively studied due to their crucial roles in pathological processes. On the basis of experimental studies published during the last decade in relation to antioxidant, anti-inflammatory, anti-ulcerogenic, hypolipidemic, anti-neoplastic, anti-diabetic, and anti-allergic phenomena, these bioactive phytochemicals are reviewed in this paper. Particularly, in vivo and in vitro studies have clarified that rice bran phytosteryl ferulates mediate anti-inflammatory effects by down-regulating the inflammatory transcription factor, nuclear factor κB (NF-κB), which in turn reduces expression of inflammatory enzymes such as COX-2 and iNOS, and proinflammatory cytokines such as IL-1β, IL-6 and TNF-α. Moreover, rice bran phytosteryl ferulates up-regulate blood adiponectin levels via indirect activation of peroxisomal proliferator-activated receptor γ (PPARγ) through NF-κB inhibition. In this review, we discuss potential pharmacological aspects of rice bran phytosteryl ferulates in the clinical setting.

α-mangostin, γ-mangostin, and xanthone are some of the marker compounds found in mangosteen (Garcinia mangostana Linn.) whose activity on several treatment targets including toward the peroxisome proliferator-activated receptor gamma (PPAR-γ) receptors, diphenyl peptidase 4 (DPP-4) enzyme, and aldose reductase enzyme is unknown. Although this plant has been predicted to be used as an alternative antidiabetic treatment, it has been proven through several previous studies. This research study used three natural ligands (α-mangostin, γ-mangostin, and xanthone) whose training set was designed using Molecular Operating Environment and then compared them with several drugs on the market that are used in the treatment of diabetes mellitus. The docking molecular results showed that the α-mangostin and γ-mangostin compounds had activity toward PPAR-γ receptor, DPP-4 enzyme, and aldose reductase enzyme by showing almost similar affinity values when compared to the comparison ligands. Meanwhile, xanthone showed unfavorable results. This approach shows that α-mangostin and γ-mangostin are predicted to play a role as antidiabetic mellitus in mangosteen when viewed from these mechanisms.

Diabetic kidney disease (DKD) is a major feature of the final stage of nearly all cause types of diabetes mellitus (DM). To date, few safe and effective drugs are available to treat. Peroxisome proliferator-activated receptors (PPARs), comprised of three members: PPAR-α, PPAR-δ and PPAR-γ, play a protective role in the DKD through glycemic control and lipid metabolism, whereas systemic activation of PPAR-γ causes serious side-effects in clinical trials. GFT505 is a dual PPAR-α/δ agonist, and the selectivity against PPAR-γ is still to be improved. Sulfuretin has been shown to suppress the expression of PPAR-γ and improve the pathogenesis of diabetic complications. In this study, by hybridizing the carboxylic acid of GFT505 and the parent nucleus of sulfuretin, we pioneeringly designed and synthetized a series of novel dual PPAR-α/δ agonists, expecting to provide a better benefit/risk ratio for PPARs. Of all the synthesized compounds, compound 12 was identified with highly activity on PPAR-α/δ and higher selectivity against PPAR-γ than that of GFT505 (EC50: hPPAR-α: 0.26 μM vs.0.76 μM; hPPAR-δ: 0.50 μM vs.0.73 μM; hPPAR-γ: 4.22 μM vs.2.79 μM). The molecular docking studies also depicted good binding affinity of compound 12 for PPAR-α and PPAR-δ compared to GFT505. Furthermore, compound 12 exhibited an evidently renoprotective effect on the DKD through inhibiting

inflammatory process, which might at least partly via JNK/NF-κB pathways in vivo and in vitro. Overall, compound 12 hold therapeutic promise for DKD.

Catalpa pod has been used in traditional medicine for the treatment of diabetes mellitus in South America. Studies on the constituents of Catalpa species have shown that it is rich in iridoids. In the present study, three previously undescribed compounds (2-4), including two secoiridoid derivatives along with twelve known compounds, were isolated from the fruits of Catalpa bignonioides Walt. In addition, fully assigned 13C-NMR of 5,6-dihydroxy-7,4'-dimethoxyflavone-6-O-sophoroside (1) is reported for the first time in the present study. The structures of compounds were determined on the basis of extensive spectroscopic methods, including UV, IR, 1D, and 2D NMR, mass spectroscopy, and CD spectroscopic data. All the isolated compounds were evaluated for α-glucosidase inhibitory activity. Among the tested compounds, compounds 2, 3, and 9 exhibited significant inhibitory activity against α-glucosidase enzyme assay. Meanwhile, the effect of compounds 2, 3, and 9 on glucose-stimulated insulin secretion (GSIS) was measured using pancreatic β-cells. Compounds 2, 3, and 9 exhibited non-cytotoxicity-stimulated insulin secretion in INS-1 cells. The expression levels of proteins associated with β-cell function and insulin secretion such as phosphorylation of total insulin receptor substrate-2 (IRS-2), phosphatidylinositol 3-kinase (PI3K), Akt, activated pancreatic duodenal homeobox-1 (PDX-1), and peroxisome proliferator-activated receptor-γ (PPAR-γ) were increased in INS-1 cells after treatment with compounds 2, 3, and 9. The findings of the present study could provide a scientific warrant for their application as a potential antidiabetic agent

Experimental Work

The human PPAR-gamma receptor

The Protein Data Bank (PDB) database (https://www.rcsb.org/) was searched with keyword "PPAR" resulting 424 reported crystal structures. The result was customized for human specific entries thus resulting 407 crystal structures. After searching in these 407 entries the peroxisome proliferator-activated receptor gamma (PPARG) was selected with (PDB ID : 4PRG). It's a 270 amino acid residue long protein with a molecular weight of 125.79 kDa. The crystal structure was determined by X-ray diffraction method with a resolution of 2.90 Å. There were four chains (A, B, C and D) with a co-crystal agonist (PDB ID: 072). The structure was downloaded in PDB format for further investigation.

4.1.2 Selection of natural compounds from PubChem

The NCBI PubChem database (https://pubchem.ncbi.nlm.nih.gov/) was searched with the common name of the reported natural compound agonists as keyword and the corresponding compounds were downloaded in SDF format for further investigation. A list of physio-chemical properties of ten natural compound agonists is given in table-1 and the their 3D structural views are shown in figure-1.

Table-1: The physio-chemical properties of selected natural compound agonists

Sl. No.	Compound name	PubChem CID	Molecular Weight	XLogP	Hydrogen bond donor count	Hydrogen bond acceptor count
1	Amorphastilbol	6440462	348.500	7.3	2	2
2	Biochanin_A	5280373	284.260	3.0	2	5
3	Catechin	73160	290.270	0.4	5	6
4	Genistein	5280961	270.240	2.7	3	5
5	Honokiol	72303	266.300	5.0	2	2
6	Kaempferol	5280863	286.240	1.9	4	6
7	Luteolin	5280445	286.240	1.4	4	6
8	Magnolol	72300	266.300	5.0	2	2
9	Quercetin	5280343	302.230	1.5	5	7
10	Resveratrol	445154	228.240	3.1	3	3

Methods

4.2.1 Molecular docking studies of natural compounds on human PPAR-gamma receptor

4.2.1.1 Preparation of natural compounds

All ten natural compounds (6440462, 5280373, 73160, 5280961, 72303, 5280863, 5280445, 72300, 5280343 and 445154) were prepared in AutoDockTools by opening their .pdb format files individually. During preparation the compounds were added with Gasteiger charges, polar hydrogen and selecting all resonable rotatable bonds. The selection of rotatable bonds is important as it makes the compounds flexible so that it can easily accommodated in the receptor active sites. Then they were saved in .pdbqt format for further docking studies in AutoDock VINA.

4.2.1.2 Preparation of PPAR-gamma receptor models

The human PPAR-gamma receptor model (PDB ID: 4PRG) was imported to AutoDockTools in .pdb format and prepared for docking. During preparation the receptor models were added with the polar hydrogens and saved in .pdbqt format for further analysis.

4.2.1.3 Preparation of grid box for docking

Prior to preparation of grid box the receptor structure 4PRG was analyzed for the selection of reported co-crystal ligand 072. The amino acid residues lining 5Å area of the 072 were selected for defining the active site as shown in figure-2. The selected residues were fetched in the prepared receptor and the grid box was prepared containing these residues (figure-3). After preparation of grid box the grid parameters were saved for their inclusion in the configuration file.

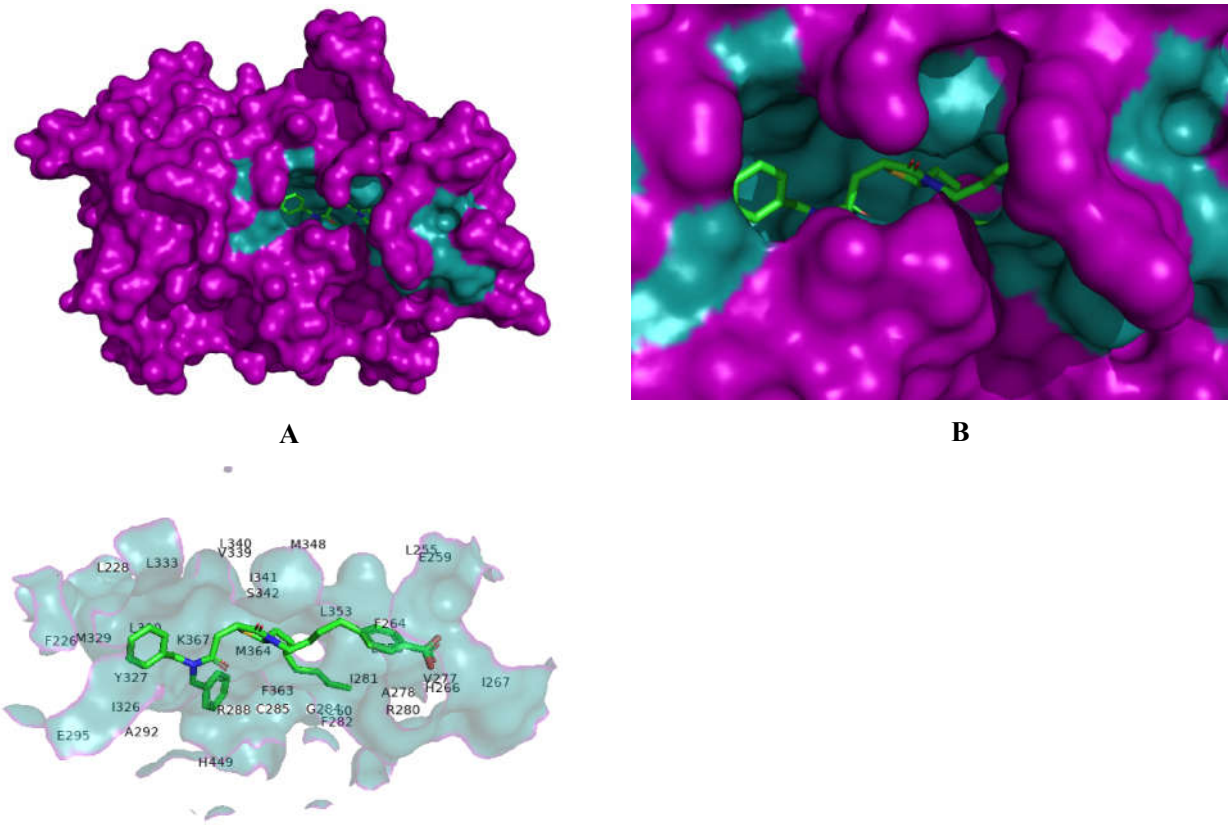


Figure-1: A: surface view of the active site containing 072, B: Close view of the active site with co-crystal ligand 072 C: Amino acid residues lining 5 Å area of 072

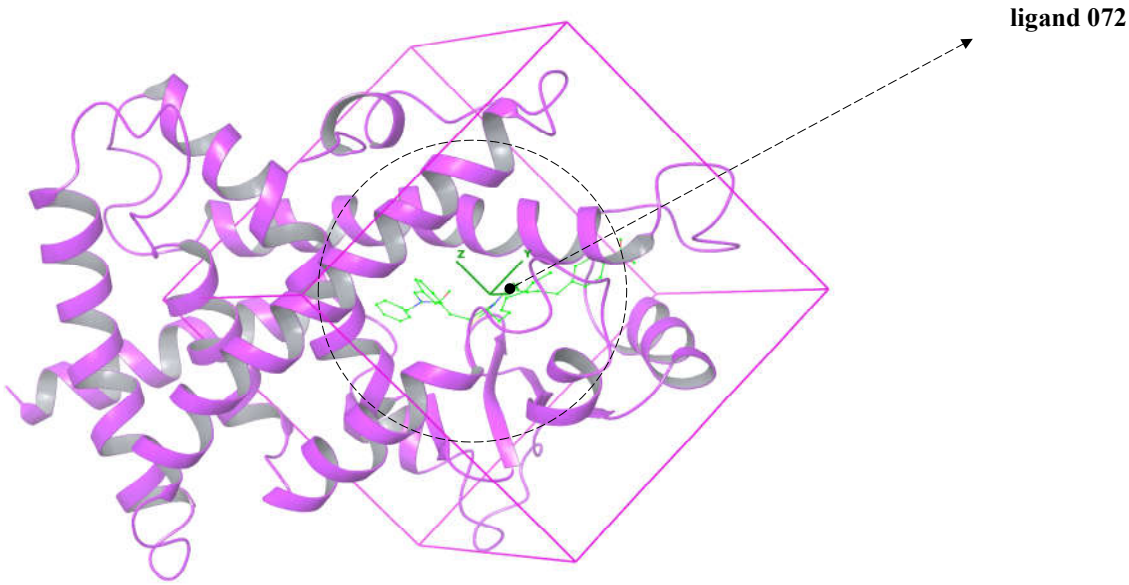


Figure-2: The selected receptor with the grid box containing 072.

Docking of natural compounds on the receptor

All ten natural compounds were docked in the prepared receptor model of human PPAR-gamma by specifying their information in the configuration files containing grid parameter files and other parameters like energy_range set to 4 and exhaustiveness set to 9. About 10 docking runs were done in AutoDock VINA by specifying the configuration files and generating docking poses in out.pdbqt files whole docking result in log.txt files. A sample configuration file information is shown below.

receptor = pparg_no_ligand.pdbqt

ligand = 72300.pdbqt

center_x = 17.55

center_y = 64.14

center_z = 10.23

size_x = 30

size_y = 30

size_z = 30

energy_range = 4

exhaustiveness = 9

Results and Discussions

Results

Molecular docking of natural compound agonists on PPAR-gamma receptor

The molecular docking studies of ten natural compound agonists on PPAR-gamma receptor were analyzed for selection of best compound that can act as a suitable agonist in future. The post docking analysis revealed that each compound has about 9 conformation poses with some reasonable readings of docking scores. The docking analysis was further refined by selecting the highest scored conformation poses of all ten compounds on PPAR-gamma and receptor. It was observed that compound1 (Amorphastilbol) had highest score (-9.3) and compound10 (Resveratrol) had lowest score (-7.8). These observations are mentioned highlighted in table-2 and the surface representation of best docking poses shown in figure-4, figure-5 and figure-6.

Table-2: Post docking analysis of all ten natural compounds on PPAR-gamma receptor

Sl. No.	Compound name	PubChem CID	Receptor PDB ID	Docking score
1	Amorphastilbol	6440462	4PRG	-9.3
2	Biochanin_A	5280373	4PRG	-8.0
3	Catechin	73160	4PRG	-8.4
4	Genistein	5280961	4PRG	-8.4
5	Honokiol	72303	4PRG	-8.4
6	Kaempferol	5280863	4PRG	-8.3
7	Luteolin	5280445	4PRG	-8.4
8	Magnolol	72300	4PRG	-8.1

9	Quercetin	5280343	4PRG	-8.4
10	Resveratrol	445154	4PRG	-7.8

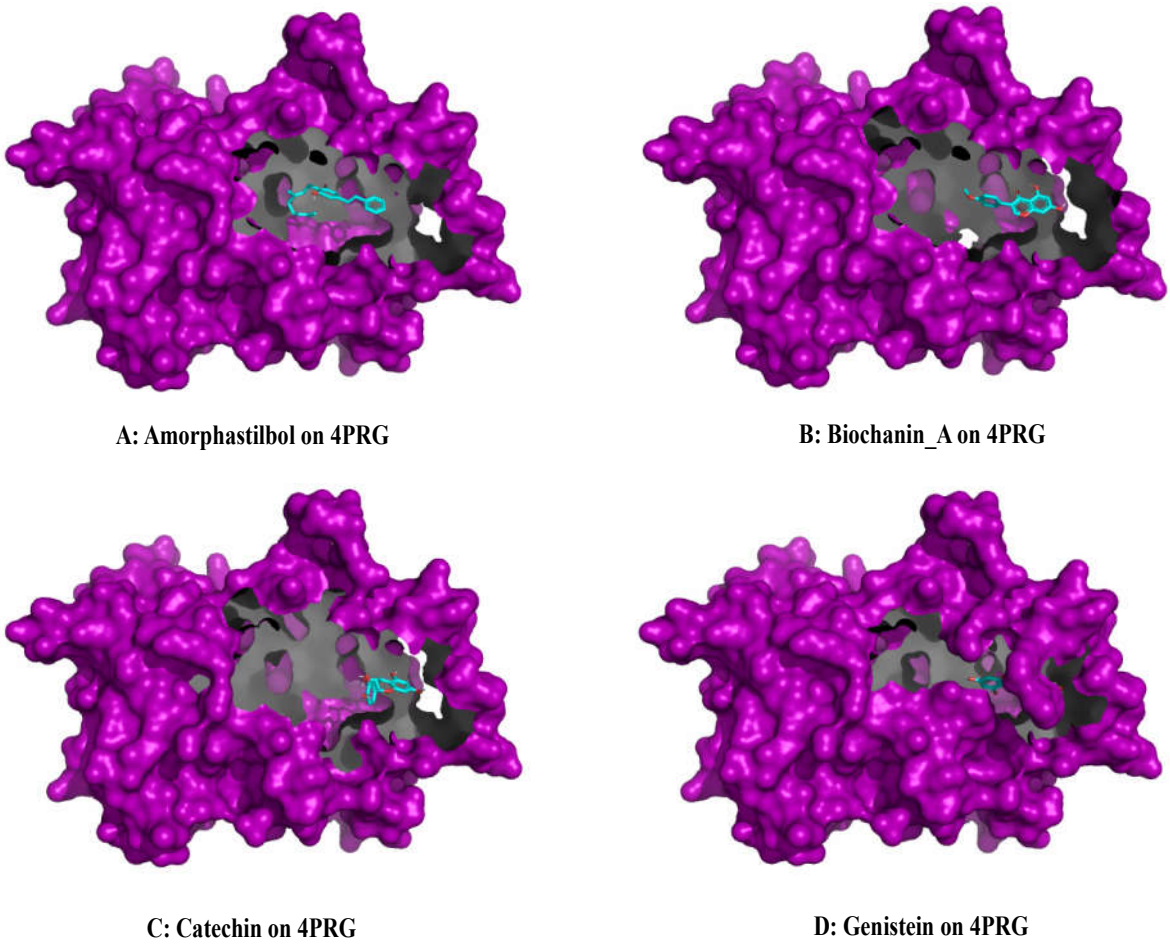


Figure-4: Surface view of best docking poses based on docking scores A: Compound1 (Amorphastilbol) on PPAR-gamma receptor, B: Compound2 (Biochanin_A) on PPAR-gamma receptor C: Compound3 (Catechin) on PPAR-gamma receptor and D: Compound4 (Genistein) on PPAR-gamma receptor.

Discussions

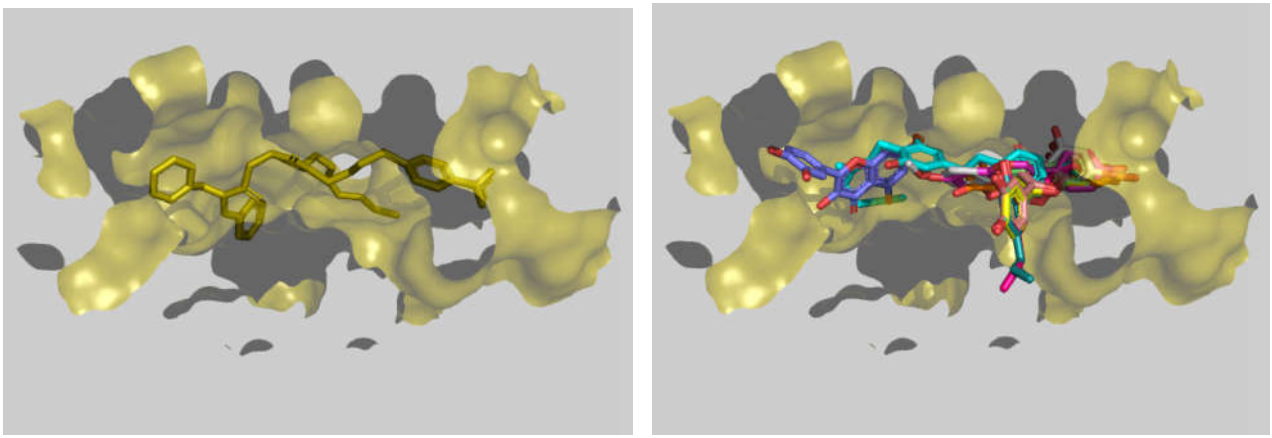
5.2.1 Molecular docking of natural compound agonists on PPAR-gamma receptor

The docking analysis is not confined to achievement of suitable docking scores by different conformation poses rather it must show the number of hydrogen formed between the residue atoms and their corresponding ligand atoms. It also important to mention the number of hydrophobic residues lining the active site where the conformation poses are accommodated.

Out of 90 conformation poses of ten natural compounds on PPAR-gamma receptor 68 poses were observed to have at least one hydrogen bond with the residues lying in the active site. The residues involved in such hydrogen bond formation were E259, E291, E295, E343, G284, G344, H226, I281, I326, K263, K280, L228, L330, L340, Q295, R228, R268 and R280.

Table-3: Hydrogen bond interaction of ten natural compounds on PPAR-gamma receptor

Sl. No.	Compound PubChem CID	Pose No.	Docking Score	Residue Atom	Ligand Atom	H-bond in A
1	72300	5	-7.5	S342:N	O2	3.2
		6	-7.5	S342:N	O1	2.9
		8	-7.4	E343:N	O2	2.8
				G344:N	O2	3.3
2	72303	1	-8.4	K263:O	H13	2.3
		2	-8.2	K263:NZ	O2	3.5
		3	-7.8	L228:N	O2	3.0
				L228:O	H14	2.8
		4	-7.7	H266:NF3	O1	3.0
		5	-7.7	E343:N	O1	3.0
		6	-7.6	I326:O	H13	2.5
				Q295:OE1	H14	2.6
		7	-7.6	E295:OE1	H14	2.2
		8	-7.5	I326:O	H14	2.3
		9	-7.5	L340:O	H13	2.5
3	73160	1	-8.4	R280:NH2	O4	3.4
		2	-8.3	S342:N	O3	3.0
		3	-8.3	L228:N	O6	3.2
				E295:OE1	H13	2.5
				E295:OE2	H13	2.9
		4	-8.2	R268:NH2	O4	3.5
		6	-8	S342:N	O2	2.9
				L330:N	O4	3.1
				L228:N	O3	3.2
		7	-7.9	L228:N	O3	3.2
		8	-7.9	G284:O	H7	2.4
				L340:O	H13	2.2
		9	-7.9	E295:OE1	H13	2.3
				E295:OE1	H14	2
				E343:N	O2	3.2
				S342:N	O3	3.1
4	445154	1	-7.8	E259:OE1	H12	2.3
		3	-7.3	R280:NE	O2	3.3
		4	-7.3	L340:O	H10	2.3
		5		L228:O	H10	2.9
		9	-7.1	E343:N	O1	2.9
				G344:N	O1	3.1



A **B**

Figure-8: Comparison of co-crystal agonist and natural compound agonists orientations in selected PPAR-gamma receptor A: Co-crystal agonist and B: selected natural compound agonists.

Summary

Diabetes mellitus seems to be a complex metabolic disorder due to poor insulin secretion which regulates the blood sugar level and maintains stability in the internal environment. Biological studies reveal that a suitable regulation of this hormone may solve the cause of this metabolic disorder to a greater extent. The human PPAR-gamma receptor is essential for adipocyte differentiation and hypertrophy and mediates the activity of insulin sensitization thus regulating diabetes mellitus to a greater extent. Several agonists are reported to date for modulation of PPAR-gamma activity which leads to the control of blood glucose levels. These are mostly chemical compounds which also induce some adverse effects besides their regulatory activities. In the current study, we report some of the reported natural compounds as a potential agonists of human peroxisome using computational techniques. About ten natural compounds were docked in the active site of the human PPAR-gamma receptor (PDB ID: 4PRG) with a co-crystal ligand (PDB ID:072). The post docking analysis revealed that most of the natural compounds occupied the same active site defined by the co-crystal ligand with some reasonable docking scores reaching (>9.0 kcal/mol) and important interaction with some conserved residues of the receptor.

Conclusion

In the current work we report the molecular docking of ten natural compound agonists of human PPAR-gamma receptor. Although there are many small molecular agonists are reported and used for the regulation of PPAR-gamma receptor they are associated with many worst chemical side effects. We also report about ten natural compounds as potential agonists for human PPAR-gamma receptor in the light of many agonists are reported in literature and used the same binding site reported for agonists (like 072). Our finding are quite generous and we found almost all natural compound agonists suitable to be accommodated in the designated active sites of the receptor with good interactions with some conserved amino acid residues.

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