IOSR Journal Of Pharmacy And Biological Sciences (IOSR-JPBS)
e-ISSN:2278-3008, p-ISSN:2319-7676. Volume 15, Issue 4 Ser. II (Jul. –Aug. 2020), PP 59-73
www.Iosrjournals.Org

Analysis of New Potent Anti-Diabetic Molecules from Phytochemicals of *PistiaStrateotes* with Sglt1 and G6pc Proteins of *Homo Sapiens* For Treatment Of Diabetes Mellitus. An *In Silico*Approach

*****Mudassir Lawal[1]**,** Abhishek Kumar Verma[1], Ismaila A. Umar[3], Aisha Muhammad Gadanya[4] , Bashir Umar[1], NajibLawan Yahaya[2], Bello Auwal Balarabe[1], ZainabAbdullahi Usman[1], MaimunaAliyu Adam[1],

[1]*Department Of Biochemistry, Mewar University, Gangrar ,Chittorgarh, Rajasthan (India)*
[2]*Department Of Biotechnology, MewarUniversity, Gangrar ,Chittorgarh, Rajasthan (India)*
[3]*Department of Biochemistry, Ahmadu Bello University Zaria, (Nigeria).*
[4]*Department of Biochemistry Bayero University Kano, (Nigeria)*
Corresponding Author: MudassirLawal
E mail: Mudassirlawal33@gmail.com

Abstract:
Background: *Diabetes mellitus is a chronic metabolic disorder affecting many people all over the world. The disease is associated with long-term dysfunction, failure and damage of various organs thus, affects virtually every physiological system of the body. The chronic insulin resistance, progressive decline in β-cell function or increased rate of cell death results decreased insulin production and finally leads the disease. Some therapeutic plant have showed hypoglycemic activities but the precise mechanism of action of these drugs at cellular level is yet not known and thus no better formulation of indigenous medicine could be developed till date for the treatment of the disease. In this study, we targeted enzymes (G6PC), and transporter (SGLT-1) that involved in Diabetes mellitus, we studied molecular interactions of 19 bioactive compounds in PistiaStratiotes leaves against diabetic targets namely: Glucose-6-phosphatase (G6PC, PDB ID: 1VNF) and Sodium-Glucose transporter-1 (SGLT1, PDB ID: 3DH4) were assessed.*
Material and Methods: *Molecular docking studies were performed using screening tool AutoDock 4.2.6 and the program PyRx v 0.8 docking softwares respectively.Pharmacophore analysis andthe PDB structures of bioactive compounds, Enzymes and the Enzyme-ligand interaction were visualized using Discovery Studio 4.5 and PyMOL Molecular Graphics System 1.3. The Swiss ADME was used to assess other physiochemical properties of these hit compounds*
Result: *The docking studies of multifarious ligands with the target proteins showed good inhibitory activity, amongst the compounds screenedStigmasterol (Binding energy; 3DH4: -9.7 kcal/mol, 1VNF: -6.9 kcal/mol), Phytol acetate (Binding energy; 3DH4: -9.1 kcal/mol and 1VNF: -7.2 kcal/mol), Tetracosahexaene(Binding energy; 3DH4: -8.4 kcal/mol and 1VNF: -7.3 kcal/molGlycerol-1-Palmitate (Binding energy; 3DH4: -7.4 kcal/mol and 1VNF: -6.9 kcal/mol), Diisooctyl phthalate (Binding energy; 3DH4: -7.4 kcal/mol and 1VNF: -8.6 kcal/mol). Stigmasterol, Tetracosahexaene, PhytolacetateandGlycerol-1-Palmitate had shown maximum inhibition for SGLT1(3DH4) protein whereas Diisooctyl phthalate shown maximum inhibition for G6PC (1VNF) protein. ADME/T analysis shown in Table 5, the selected properties are known to influence metabolism, cell permeation, and bioavailability. Predicted properties of Stigmasterol and Glycerol-1-Palmitate Diisooctyl phthalate Phytol acetate were within the range for satisfying all the Lipinski's rule of five to be considered as drug like potential. But on the other hand Tetracosahexaene was not satisfying all the Lipinski's rule of five to be considered as drug like potential. Under the Rule of Five, a molecule can only be orally active/absorb if it does not violate any two or more of the above rules. However, some complicated natural products are not suited to the rules.*
Conclusion: *All these four phytocompounds (Stigmasterol, Glycerol-1-Palmitate, Diisooctyl phthalate and Phytol acetate) can act as potent antidiabetic agents. They can be further subjected to fractionation and isolation to confirm their activity towards in vitro and in vivo studies and can be commercialized as a potent antidiabetic agents.*

Keywords: Molecular docking, SGLT1, G6PC, Anti-diabetic compounds

---

Date of Submission: 13-07-2020 Date of Acceptance: 28-07-2020

I. Introduction

Diabetes is a serious, chronic disease that occurs either when the pancreas does not produce enough insulin (a hormone that regulates blood glucose), or when the body cannot effectively use the insulin it produces (WHO, 2016). Raised blood glucose, a common effect of uncontrolled diabetes, may, over time, lead to serious damage to the heart, blood vessels, eyes, kidneys and nerves. More than 400 million people live with diabetes.Based on its etiology and pathology, the disease is generally classified into type-1 diabetes (T1D) and type-2 diabetes (T2D). While patients with T1D show insulin deficiency because of beta cells destruction, T2D is associated with a progressive loss of insulin secretion based on the background of insulin resistance (American Diabetes Association, 2017). In both types of diabetes, long-term hyperglycemia leads to multiple functional and structural abnormalities which damage to several organs including the kidneys, heart, eyes, nerves, and lower extremity (Lotfy *et al.*, 2016).

Mankind through observation and experience developed knowledge of the properties of plants as a source of food and medicines. Phytochemicals are as important as synthetic medicines since in some regions it is the only source of medicine. In the history of ancient civilizations, the use of medicinal herbs for curing diseases has been documented. Drugs were used in crude forms as decoctions, infusions, tinctures and poultices. Phytochemicals play an important role in the pharmaceutical industry as raw materials or as a particular drug. Secondary metabolites obtained from the plants are found to be an important source of various phytochemicals that could be used for the production of pharmaceuticals. In the developing countries, approximately 80% of the populations still rely on the traditional medicine derived from the plants for health care needs. Thus the demand for herbal medicines is continuously increasing day by day in comparison to the synthetic drugs. India is called the botanical garden of the world for its rich natural resources (Tulika & Mala, 2014).

Herbal plants are being used as medicine from ancient age and their usefulness has been recorded in human history. Herbal plants are reported to be excellent sources of several nutrients (Musa, 2005). The use of herbal drugs in treatment of diseases is found among all sections of people in India. The plant *Pistiastratiotes*, commonly known as Water cabbage or Water lettuce, as "Kainuwa" in Hausa and "Jalkumbhi" in Hindi, belongs to the family *Araceae*, is an edible, aquatic, floating ornamental plant widely distributed across tropical and sub-tropical areas around the world. The plant's leaves are light green, obovate with prominent longitudinal veins at its base (Arber, 1991).

*P.stratiotes*plantextracts have been shown to contain various alkaloids, glycosides, flavonoids and phytosterols (Khare, 2005). It has been used traditionally in the treatment of various diseases such as diabetes, eczema, leprosy, ulcers, piles, stomach disorder, throat and mouth inflammation, to mention a few (Khan *et al*., 2014). Previous studies have shown that various extracts from *P. stratiotes*possessed anti-inflammatory (Koffuor*et al*., 2012), antioxidant (Jha*et al.*, 2010), antimicrobial and antidiarrheal activities (Rahman*et al*., 2011). Now, the antidiabetic potential of *P. stratiotes*has been adequately documented (Lawal*et al.,* 2019).

The chemical composition analyzed by GC–MS from areal part of *P. stratiotes*suggested five main families: tannins, terpenoid, glycosides, flavonoids and phytosterols which are strong anti-oxidant compounds (IJBMS, 2011). Possessing polyphenol structure involving high number of hydroxyl group inside, tannins and flavonoids were thus, predicted to be able to form hydrogen bonds with various reactive oxygen species, such as singlet oxygen, peroxynitrite and hydrogen peroxide which are major causes of cell damages. Due to this mechanism, tannins and flavonoids were considered to play potential roles in reducing the oxidative stress related to Type 2 DM (Evans, 2007). Terpenoids are a large class of organic compounds in plants whose potential antioxidant activity has been confirmed (Gonzalez-Burgos and Gomez-Serranillos, 2012).

However, there are no researches indicating the affinity of bioactive compounds of *P. stratiotes* on 'druggable' targets in both Type 1 and Type 2 Diabetes mellitus. The two (2) targeted proteins used in this study were previously investigated to serve as potential drug target for Type 1 and Type 2 DM (Nguyen and Le, 2012; Shi, 2009).

In humans with type 2 diabetes mellitus and in hyperglycemic mice, the renal SGLT-2 expression is increased. SGLT-2 inhibitors are effective in lowering blood glucose when used as monotherapy or in combination with other oral agents/ insulin. These results have been documented in large multicenter studies. Metformin sometimes causes intolerable gastrointestinal side effects in several patients, though it is the first choice for treating type 2 diabetes mellitus. SGLT-2 inhibitors apart from producing glycosuria, seem to slow the progression of diabetic nephropathy by means of hemodynamic changes as well as by the tubule-glomerular feedback. Moreover, SGLT-2 inhibitors comprise a new class of glucose-lowering drugs that provide small reductions in body weight and blood pressure(Deepak *et al.*, 2018).

Glucose-6-phosphatase (G-6-Pase), which is predominantly, located in the liver, catalyses the terminal step in both gluconeogenesis and glycogenolysis by converting glucose-6-phosphate, (G-6-P), to glucose and inorganic phosphate making it a key regulating step in blood glucose homeostasis. This has been demonstrated in patients with glycogen storage disease Type, where the enzyme is deficient, causing, amongst many clinical features, hypoglycaemia. In contrast, the catalytic enzyme activity, protein content and mRNA levels are all

increased in diabetic animals and presumably contribute to the prevailing hyperglycaemia. All these features can be reversed by administration of insulin (Madsen &Westergaard, 2001).

Besides playing a central role in hepatic glucose production, the augmented G-6-Pase activity in β-cells could also play a pivotal role in the impaired insulin secretion seen in Type 2 diabetes. Khan and co-workers have recently looked at the effects of various G-6-Pase inhibitors, including the T1-translocase inhibitor on islet and hepatic G-6-Pase activity in ob/ob mice. Thus, any compound that inhibits hepatic and/or islet G-6-Pase activity could be beneficial as an antidiabetic agent via decreased hepatic glucose production and/or stimulated insulin release leading to improved plasma glucose levels(Madsen & Westergaard, 2001). The Objectives are to predict the binding affinities of different bioactive compounds and to illustrate specific areas of interaction between the ligands and the amino acid residues of a target proteins using *in silico*analysis as well as to evaluate the absorption, distribution, metabolism and excretion- toxicity (ADME-T) parameters for the identified hit compounds.

II. Material And Methods

Materials

AutoDock 4.2software which perform the automated Docking of Flexible Ligands to Flexible Receptors, introduced by Garrett M. Morris et al., popularly known as Autodock with version 4.2 was used in the present study to study the molecular docking. Discovery Studio, a BIOVIA's software of comprehensive predictive science application for the Life Sciences, was used in the present study to separate the ligand from the protein. PyMOL was used to visualize the docking positions in 3-dimensional images, it is an open a source, user-sponsored, molecular visualization system created by Warren Lyford De Lano.

Retrieval of Proteins

A- Homology Modeling

Homology modeling is probably the most effective tool for producing full-bodied tridimensional models of protein structure. Homology modeling approaches use experimental protein structure ("templates") to construct drug discovery receptor proteins. Swiss-model is a web-server of structural bioinformatics dedicated to modeling homology for the analysis f 3D protein structures (Chothia C, LesKAM, 1986; Kaczanowski, Zielenkiewicz, 2010). Human G6PC FASTA sequence (Accession ID: P35575) was retrieved from Uniprot database. The whole protein was modeled by uploading the G6PC protein FASTA sequence into the SWISS-MODEL workspace via automatic mode for the creation of a more reliable protein model. The G6PC protein and its sequence were selected as the target protein and query sequence respectively. The Vanadium Chloroperoxidase (PDB ID: 1VNF) structure that derived from a sample of proteins, served as a reference templates. The prototype, 1vnf.1.A demonstrated to query sequence the highest sequence identity, which was used to create an improved G6PC protein model. Global quality estimate, local quality estimate comparison, and G6PC model template alignment with 1vnf.1.A, were calculated. Thus, the three dimensional crystal structure of Vanadium Chloroperoxidase (PDB ID: 1VNF) was retrieved from RCSB PDB database for use as the template for validation.

B. Modeled G6PC protein validation

The structure contains a few amino acid residues that are absent. The G6PC protein and its sequence were selected as the target protein and query sequence respectively. The protein model was built using 1vnf.1.A, the protein as a template (Fig. 1). Ramachandran plot validated the G6PC protein output using Rampage (Read *et al.*, 2011) and SPDBV (Deep ViewSwiss-Pdb Viewer) version 4.10, based on the RMSD value obtained by superimposing the G6PC protein model on its 1vnf.1.A, (Savarino*et al.*, 2007) model design. The G6PC based protein plot values for Ramchandran and its 1vnf.1.A template were obtained. In the G6PC protein Ramachandran plot, 97.9 percent of the amino acid residues were found in the favored region, 1.2 percent residues in the permitted region, and 0.9 percent of the residues in the outer regions (Figure 3). For the 1vnf.1.A, the Ramachandran plot displayed 96.5 percent, 2.8 percent, and 0.7 percent, respectively, of the residues in the favored, permitted, and outer regions. The Ramachandran plot data for the G6PC and 1vnf.1.A modeled proteins suggested favorable reliability of the G6PC-modelled protein for subsequent docking studies (Figure 3).

Receptor preparation

The structural information of the macromolecules determined by x-ray crystallographic and NMR methods are available in the PDB. The 3D structure of SGLT1 and G6PC were taken from Protein Data Bank (http://www.rcsb.org/pdb/) as follow SGLT1(PDB ID: 3DH4) and G6PC(1VNF) and PTP1B(PDB ID: 4Y14). All these target structures were tested again at the binding site, in order to verify the capacity of the model in reproducing experimental observation with new ligand, Following this way, SGLT1(PDB ID: 3DH4) was tested with Sotagliflozin(standard) and G6PC(1VNF) was tested against Thielavin A (standard).

Table 1: Names of target proteins with their Protein Data Base (PDB) Identification Number

S/N	Target proteins	Abbreviation	PDB ID No.
1	Glucose-6-phosphatase	G6PC	1VNF
2	Sodium-Glucose transporter-1	SGLT1	3DH4

Active Site

The active site is predicted using PDBsum, which is a pictorial database of 3D structures in the Protein Data Bank database. The default active site was considered of docked complexes, Amino acid within 10^0A by cogitating ligand of interest in center (Rekha and Chandrashekhara, 2017).

Bioactive compounds (Ligands) preparation

The several literature searched were carried for the identification of different phytochemicals In *PistiastratiotesL*. At the end of the literature searched, we found the presence different bioactive compounds in *PistiastratiotesL* leaves and root. A total of 19 ligands of *Pistiastratiotes* were selected and used for the present study.Most of the 3D or 2D structures of phytochemicals *in P. stratiotes* were downloaded from PubChem, compound section of National Center for Biotechnology Information (NCBI) and the others were drawn by Gauss- View 5 (Denningto*net al*. 2009). The phytochemical with 2D conformation was converted to 3D conformation using open Babel molecule format converter. Marvin Sketch software (version 15.10.0) was used to perform the conversion from .sdf to .pdf (for docking) and mol (for molecular properties prediction) file. For the protein were prepared using Marvin Sketch. Ligand's energy was minimized by relating the mmff94 force field and conjugate gradients optimization algorithm using PyRx-Python prescription 0.8 for 200 steps (Dallakya*net al.,* 2015). Ligands during this process also being checked for Torsion count to detect currently active bonds with default settings. Importantly, amide bonds were checked and treated as non-rotatable using Swiss ADME.

Ligand based ADME/Toxicity prediction

ADME properties determine drug-like activity of ligand molecules based on Lipinski's rule of five. For the compound to be selected as a hit, it must be non-hepatotoxic and non-carcinogenic. Swiss ADME was used to assess other physiochemical properties of these hit compounds (Daina*et al.,* 2017). This technique has been widely used as a screen for compounds that are expected to be produced more for product design programs. We have tested parameters such as the number of rotatable No. bonds (> 10) and the number of rigid bonds that suggest good oral bioavailability and good intestinal absorption of the compound (Ertl P *et al.,*2000).

Docking-based virtual screening

The retrieved hit compounds from pervious screening was subjected to docking-based virtual screening against the 3D structure of different Target proteins (PDB ID) using AutodcokVina in PyRx 0.8 program (Dallakyan and Olson, 2015). Before docking, hit compounds were energy minimized and converted from sdf files into pdbqt files using Open Babel tool in PyRx 0.8 program (O'Boyle *et al*., 2011). The grid box was cantered to cover the amino acid residues involved in the topology of the primary pocket of different Target proteins. Prior screening, Standard inhibitor was added to the database as a control. Compounds that bind to Target protein with high binding affinities in comparison to Standard inhibitor were considered for further analysis(Verma*et al*., 2020).

Table 2: GC-MS DATA: Molecular Formula, Molecular Weight and Pubchem ID number of Bioactive Compounds of *Pistiastratiotes*Leaves of Ethanol extracts(Tyagi& Agarwal, 2017).

S/N	Name Of Compounds	Molecular Formula	Molecular Wt	Pubchem ID
1	Isobutyl alcohol	$C_4H_{10}O$	74	6560
2	Formic acid,1- methylethyl ester	$C_4H_8O_2$	88	12257
3	Propane,1,1-diethoxy-2-methyl	$C_8H_{18}O_2$	146	519415
4	L-Glutamine	$C_5H_{10}N_2O_3$	146	5961
5	n- Hexadecanoic acid	$C_{16}H_{32}O_2$	256	985
6	Hexadecanoic acid, ethyl ester	$C_{18}H_{36}O_2$	284	12366
7	Linolelaidic acid, methyl ester	$C_{19}H_{34}O_2$	294	5362793
8	9,12,15- Octadecacatrienoic acid, methyl ester, (Z,Z,Z)	$C_{19}H_{32}O_2$	294	9316
9	12,15- Octadecadiynoic acid, methyl ester	$C_{19}H_{30}O_2$	290	538453
10	Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl) ethyl ester	$C_{19}H_{38}O_4$	330	123409
11	Diisooctyl phthalate	$C_8H_4(C_8H_{17}COO)_2$	390	33934

12	Docosanoic acid, ethyl ester	$C_{24}H_{48}O_2$	368	22199
13	Stigmasterol	$C_{29}H_{48}O$	412	5280794
14	Tetradecane	$C_{14}H_{30}$	198	12389
15	Phytol, acetate	$C_{22}H_{42}O_2$	338	6428538
16	2-Hexadecan-1-0l, 3, 7,11, 15-tetramethyl (R-[R*, R*- E])-(CAS)	$C_{20}H_{40}O$	296	9018
17	Glycerol 1- Palmitate	$C_{19}H_{38}O_4$	330	14900
18	2,6,10,14,18,22 Tetracosahexaene	$C_{30}H_{50}$	410	638072
19	Trideutetro Methyl Ethyl Ether	$C_3H_5D_3O$	60	10903

Molecular docking study

Hit compounds fulfilling the pervious filters were docked against the 3D structure of different Target proteins using Autodockvina program (Trott and Olson, 2010). The protein structure (PDB: ID) was obtained from RCSB protein data bank (Villa *et al.*, 2007). Discovery studio 4.5 (Accelrys, San Diego, CA, USA) was used to remove the unwanted water molecules and ligands as well as to generate the pdb files for the protein in monomer form. Autodock tools program was used to generate the pdbqt files and to prepare the gridbox for the docking configuration files (Sanner, 1999). The content of configure file was determined as position of receptor/enzymes file, ligand file, data of Grid- box's three coordinates X, Y, Z were 19.6703, -20.881, 73.7109 respectively in case of SGLT1 and -27.333, 60.9328, -0.0814 for G6PC, the size of Grid Box which was set up in 25 X 25 X 25 points. The grid box includes the whole binding site of the proteins line and provides sufficient space for the ligands translational and rotational walk.

Pharmacophore analysis

This part of process was carried out by using the pharmacophore tool included in LigandScout (Wolber and Langer 2005). The program showed us the 2D and 3D structure with the position and interaction of ligand in the binding pocket of the receptor. From these 2D figures, some types of bond were identified by color and symbol. Four features namely hydrogen bond acceptor (HBA), hydrogen bond donor (HBD), negative ionizable area (NIA), hydrophobic interaction were labeled as red arrow, green arrow, red star and orange bubble (supporting information) respectively.

Visualization of Binding Interaction

The PDB structures of five bioactive compounds, Enzymes and the Enzyme-ligand interaction were visualized using Discovery Studio 4.5 and PyMOL Molecular Graphics System 1.3

III. Result And Discussion

Free energy binding of phytochemicals to targeted proteins

The result in (Fig. 4) showed the binding energy of all the nineteen bioactive compounds present in *P.stratiotes*on 5 target proteins related to both Type 1 and Type 2 Diabetes Mellitus in humans. Among those docking result, the complete value of binding affinities ranged from -4.8 to -11.9 (kcal/mol; Fig. 4). In this range, the greatest result was showed in five bioactive compounds which were higher standards in term of binding energy.

Those are Tetracosahexaene, Glycerol-1-Palmitate, Stigmasterol, Diisooctyl phthalate and Phytol acetate which were selected for pharmacophore analysis step. Besides that, Stigmasterol and Phytol acetate also had very good results but this was different in each protein, perhaps the amino acid construction of each protein. The binding affinities of Stigmasterol were -9.7 and -6.9 kcal/mol in SGLT1(3DH4) and G6PC(1VNF) respectively, Phytol acetate were 9.1 and 7.2 kcal/mol in SGLT1(3DH4) and G6PC(1VNF) respectively,Tetracosahexaene were -8.4, and -7.3 kcal/mol in SGLT1(3DH4) and G6PC(1VNF) respectively, Glycerol-1-Palmitate were -7.4 and -6.9 kcal/mol in SGLT1(3DH4) and G6PC(1VNF) respectively, Diisooctyl phthalate were -7.4 and -8.6 kcal/mol inSGLT1(3DH4) and G6PC(1VNF) respectively.

Also, Fig. 4 also indicated the best receptor for these bioactive compounds in *P.stratiotes*. From the result of this chart, the line for SGLT1(3DH4), stayed at the upper level and G6PC(1VNF) located at bottom of chart. This proves that SGLT1 protein was the best receptor by showing high binding affinity to the plant's bioactive compounds (Stigmasterol, Diisooctyl phthalate, Glycerol-1-Palmitate and Phytol acetate) compared to the G6PC protein which showed the lower binding affinity to the bioactive compounds (Stigmasterol, Diisooctyl phthalate, Glycerol-1-Palmitate and Phytol acetate) of *P.stratiotes.*

The line of SGLT1(3DH4) always showed in highest level as shown by the result. It means that there is stronger interaction of ligand on this protein, compared to other target protein. Furthermore, in the active site of SGLT1 and G6PC many compounds of *P.stratiotes* had stronger binding affinity than the controls and 50 % of compounds in *P.stratiotes* can interact with SGLT1by absolute value of binding energy higher 6.1 kcal/mol

(Fig: 4) and this result statistically proved that, *P.stratiotes*is potential drug for some druggable targets related to both Type 1 and Type 2 DM.

Pharmacophore analysis

SGLT1, G6PC and PTP1B

All these bioactive compounds can form either hydrophobic interaction or hydrogen bond with free residue in active site of SGLT1 protein. Tetracosahexaene, Glycerol-1-Palmitate, Stigmasterol, Diisooctyl phthalate and Phytol acetate can build up hydrogen bond with GLN284(A), VAL519(A), ASP507(C), SER453(C),LYS454(C), GLY110(A) and ILE503(C) while these amino residues residuesPRO508(C), LYS509(C), VAL280(A), PRO508(C), LYS509(C), VAL280(A), ILE107(A), PRO104(A), ILE511(A), LYS109(A), ARG121(A), PHE117(A), ILE503(C), LYS120(A), MET517(A), PRO508(C), LYS509(C), VAL280(A), ILE503(C), PRO508(C), LYS509(C), MET517(A) and VAL280(A)[Fig. 6(2, 3, 4, 5 and 6)], involved in strong hydrophobic bonds with the phytochemical. In addition, the hydrophobic interactions also played an important role in docking result. The Stigmasterol compound showed strong contact with the receptors because of the presence of two benzene rings. Stigmasterol could form not only hydrophobic interaction but also hydrogen bond with the target proteins. Furthermore, in Fig. 6(4), the residues PRO508(C), PRO508(C), ILE503(C) and VAL280(A) were frequently observed in ligand-receptor interactions between, so they could be a critical part in binding pocket.

G6PC protein formed hydrogen bonds with bioactive compounds via amino acid residues PHE407(A), PRO300(A), PHE397(A), PRO47(A), HIS404(A), TRP350(A), PRO395(A), PHE393(A), HIS496(A), PHE393(A), HIS222(A), PRO396(A), HIS38(A) while some amino acid residues in the active site of G6PC protein formed strong hydrophobic interaction with bioactive compounds, the amino acids residues are GLN220(A), HIS222(A), SER294(A), GLY293(A), ASN295(A), ARG490(A), HIS496(A), GLY403(A), HIS404(A).

ADME/T prediction of selected compound

Through using SWISSADME online tools the data was obtained and recorded in Table No.5. With the help of SWISSADME we had examined carious physical descriptors and pharmaceutically important properties for ADME/T prediction. All the selected phytochemicals showed important values for the various criteria tested and displayed strong drug-like properties based on the Lipinski's rule of five. The data obtained were within the range of values for all-natural compounds. The significance of polar surface area (PSA) suggested good oral bioavailability for natural compounds (Stigmasterol, Diisooctyl phthalate, Glycerol-1-Palmitate and Phytol acetate). The parameters, such as number of rotatable bonds and number of stable bonds correlated with the product of intestinal absorption, revealed that all-natural compounds (Stigmasterol, Diisooctyl phthalate, Glycerol-1-Palmitate and Phytol acetate) are well absorbed. All the synthesized compounds were found to be nontoxic (Table No. 5).

Online we have done ADME/T prediction, which shows our selected ligands follow all Lipinsky's rule of five. Now, we can conclude that a selected phytochemicals from *P. strateotes.*

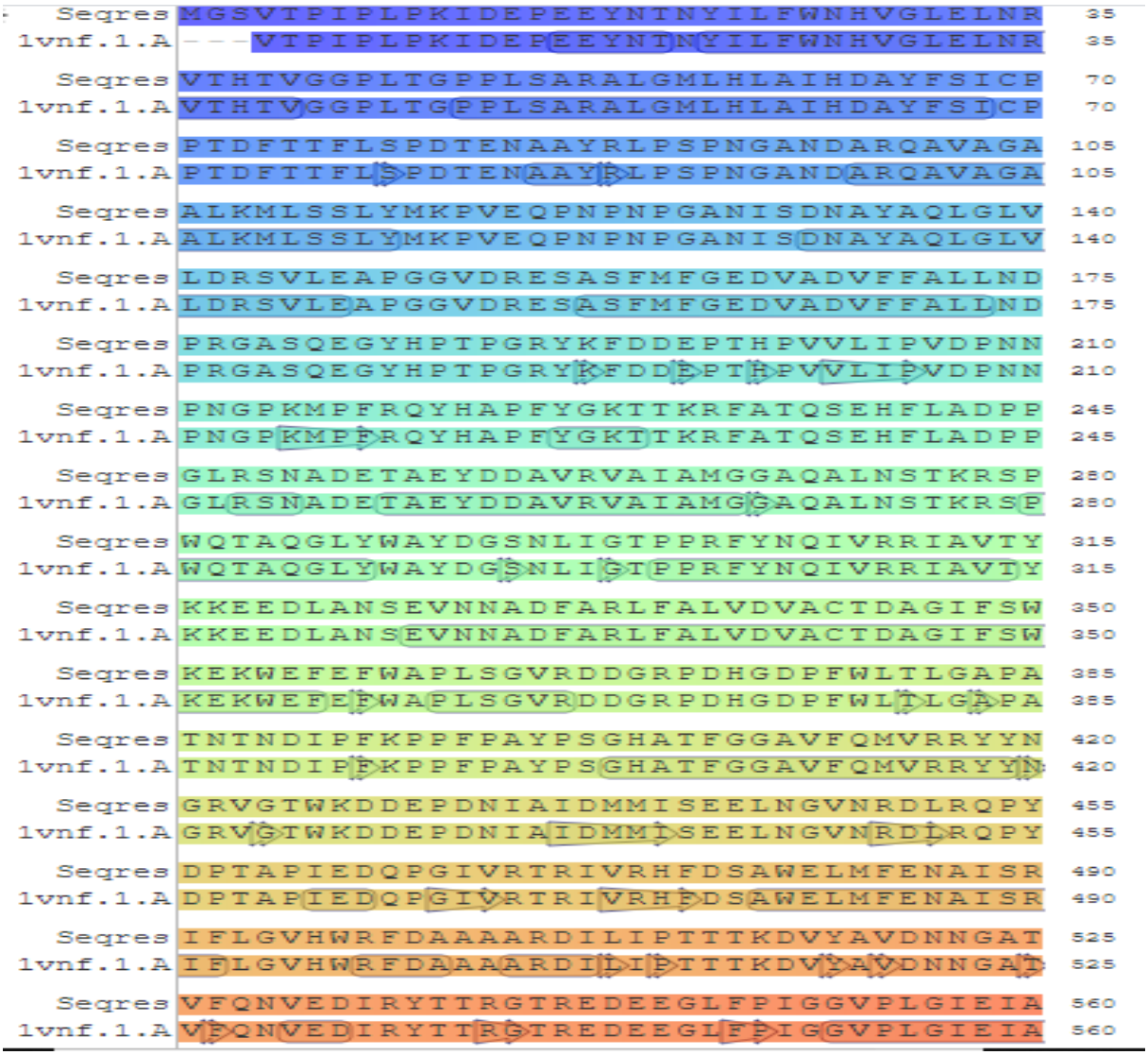


Figure 1. Aligment between Human G6PC and Vanadium Chloroperoxidase

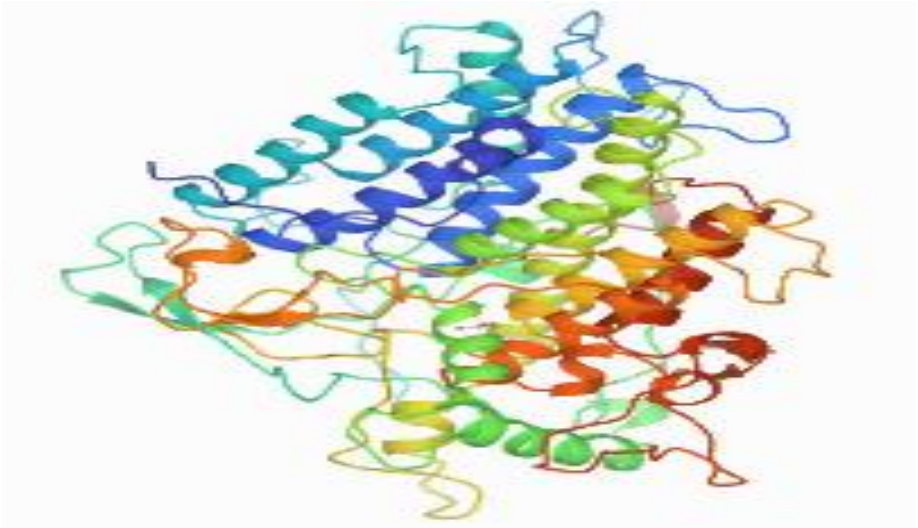
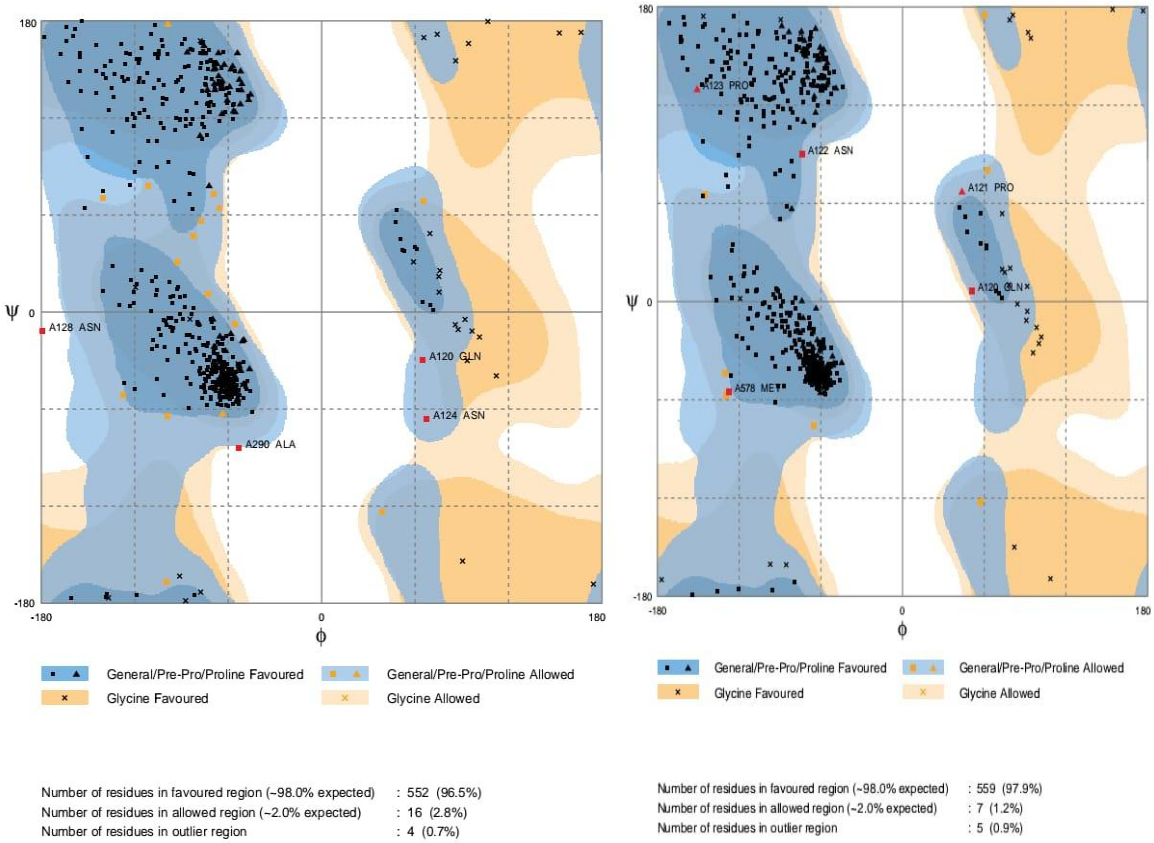


Figure 2. Homology modeled structure of Human G6PC



Number of residues in favoured region (~98.0% expected) : 552 (96.5%)
Number of residues in allowed region (~2.0% expected) : 16 (2.8%)
Number of residues in outlier region : 4 (0.7%)

Number of residues in favoured region (~98.0% expected) : 559 (97.9%)
Number of residues in allowed region (~2.0% expected) : 7 (1.2%)
Number of residues in outlier region : 5 (0.9%)

Figure 3. Ramachandran map of G6PC (Query sequence) and Template sequence model

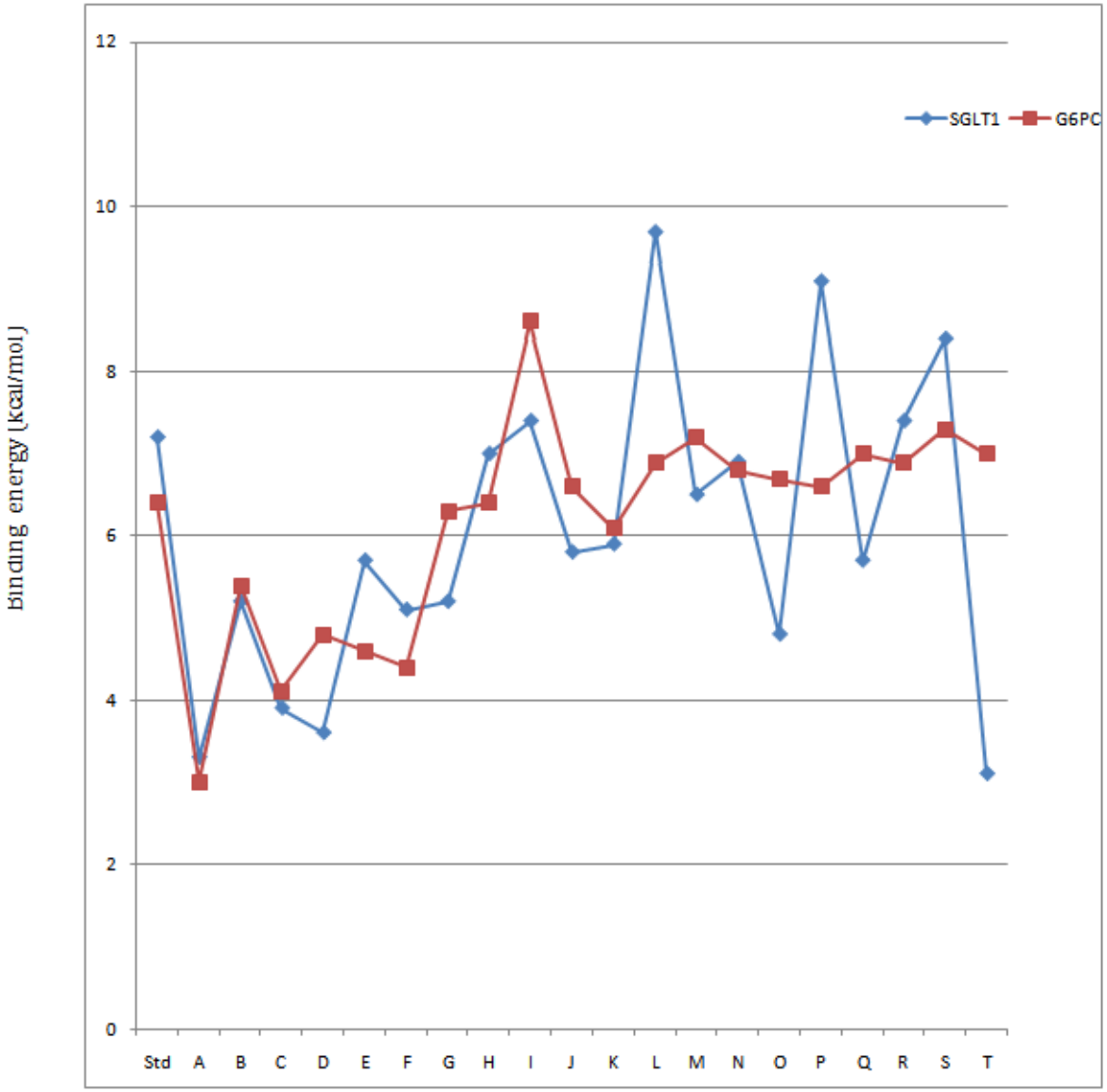


Fig 4: Standards (Acarb=Acarbose, Glim=Gliclazide, NDP=NADH Dihydro-Nicotinamide-Adenine-Dinucleotide phosphate, Stgz= Sotagliflozin and CoA=CoenzymeA)) A=Trideutetro, B=Tetradecane, C=Isobutyl alcohol, D=Formic acid,1- methylethyl ester, E =L-Glutamine, F=Propane,1,1-diethoxy-2-methyl, G =n- Hexadecanoic acid, H=Hexadecanoic acid, ethyl ester, I = Diisooctyl phthalate, J=Linolelaidic acid, methyl ester, K= Octadecacatrienoic acid, methyl ester, L=Stigmasterol, M=Octadecadiynoic acid, methyl ester, N=Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl) ethyl ester, 0=Docosanoic acid-ethyl ester, P=Phytol acetate, Q=2-Hexadecan-1-ol,3,7,11,15-tetramethyl, R=Glycerol-1-Palmitate, S=Tetracosahexaene, T=Trideutetro Methyl Ethyl Ether. Besides that, blue line represented for Alpha-Glucosidase protein, followed by the red, green, brownand light bluewere labeled for SUR-1, 11β-HSD1, SGLT1, G-6Ptase and PTP1B, respectively.

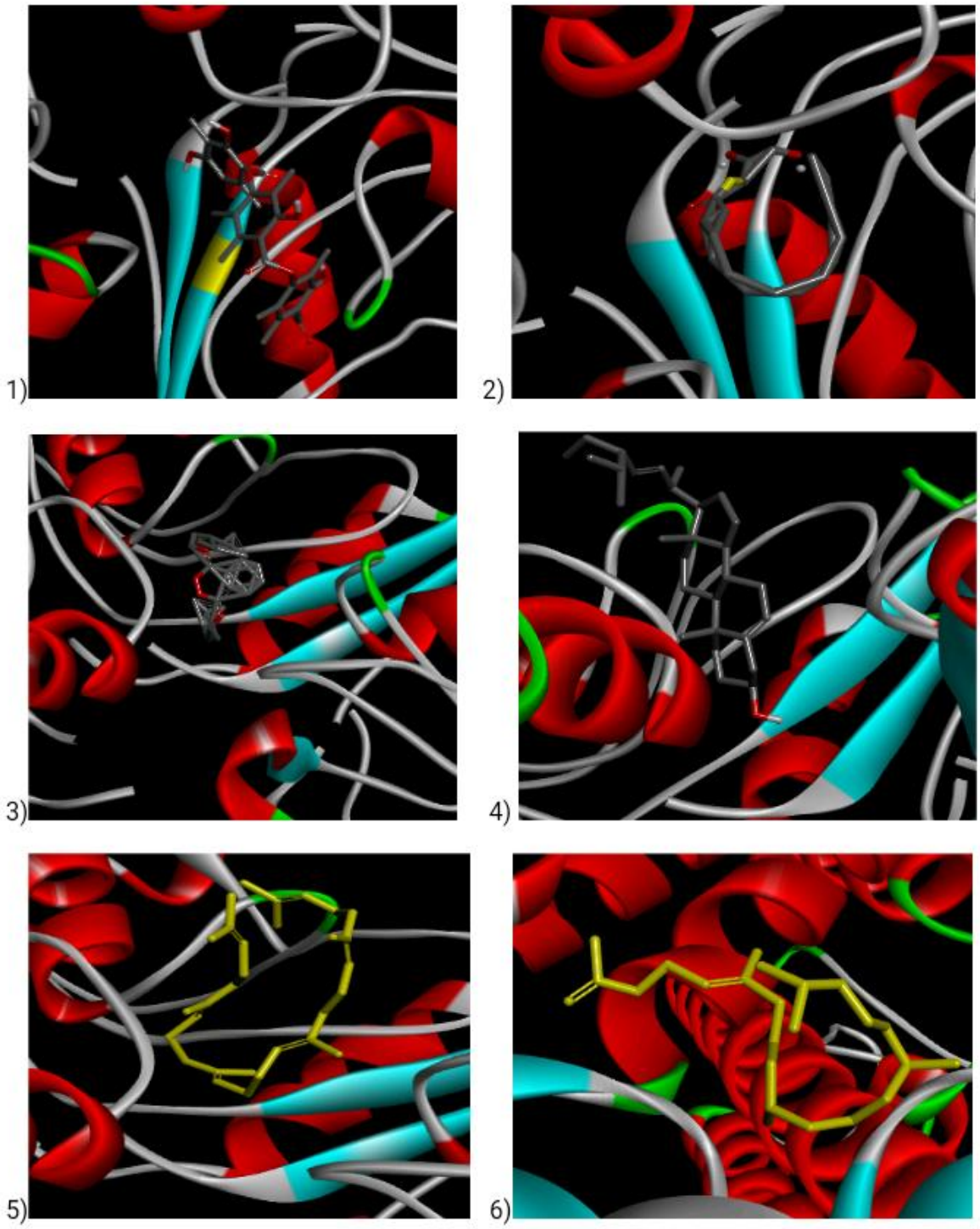


Fig 5: Ribbon representation of protein-ligand complex (1) SGLT1- Sotagliflozin (Standard), (2) SGLT1-Glycerol-1-Palmitate, (3) SGLT1-Diisooctyl phthalate, (4) SGLT1-Stigmasterol,(5) SGLT1-Tetracosahexaene and SGLT1-Phytol acetate.

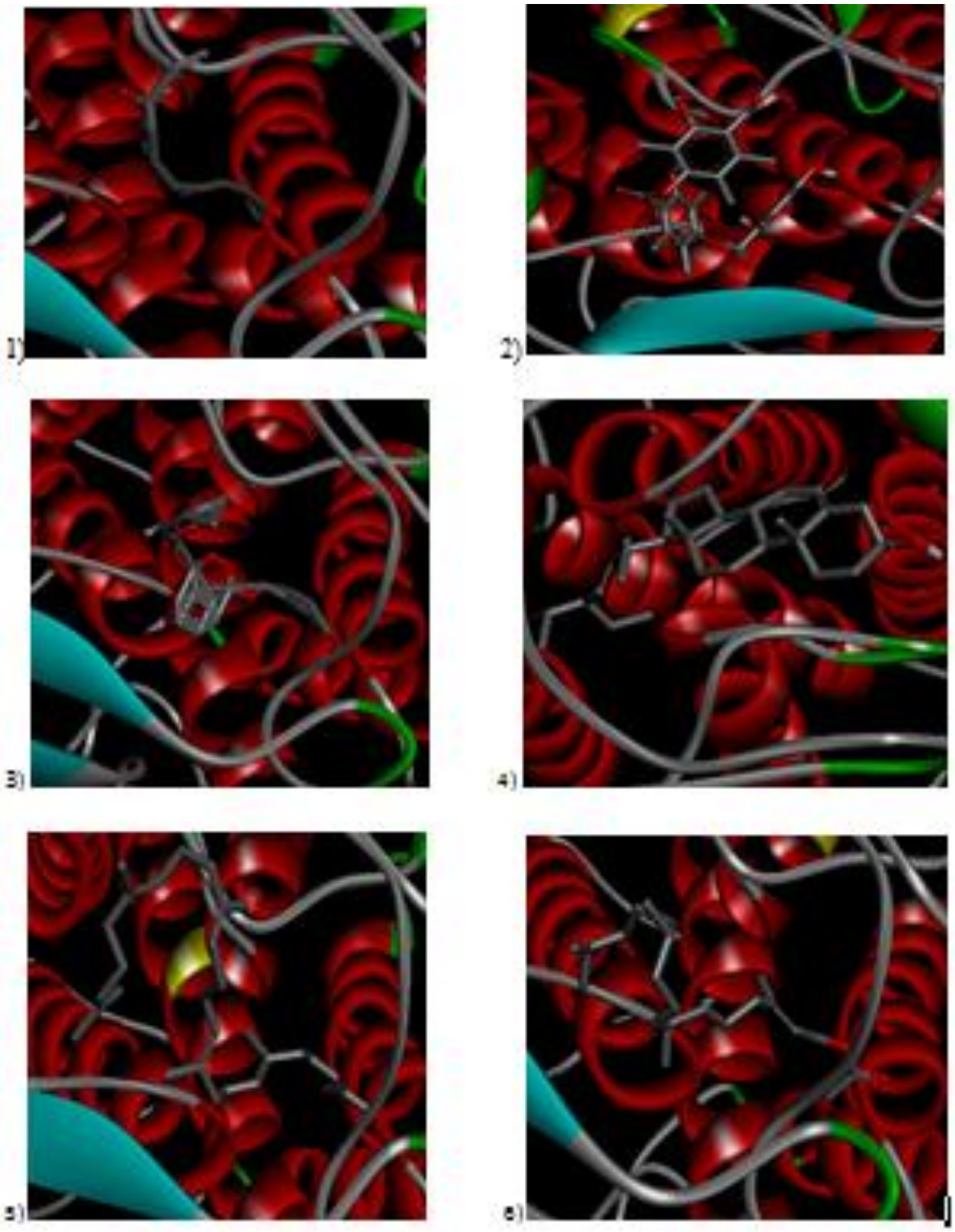


Fig 6: Ribbon representation of protein-ligand complex (1) G6PC-Thievalin A (Standard), (2) G6PC-Glycerol-1-Palmitate, (3) G6PC-Diisooctyl phthalate, (4) G6PC-Stigmasterol, (5) G6PC-Tetracosahexaene and G6PC-Phytol acetate

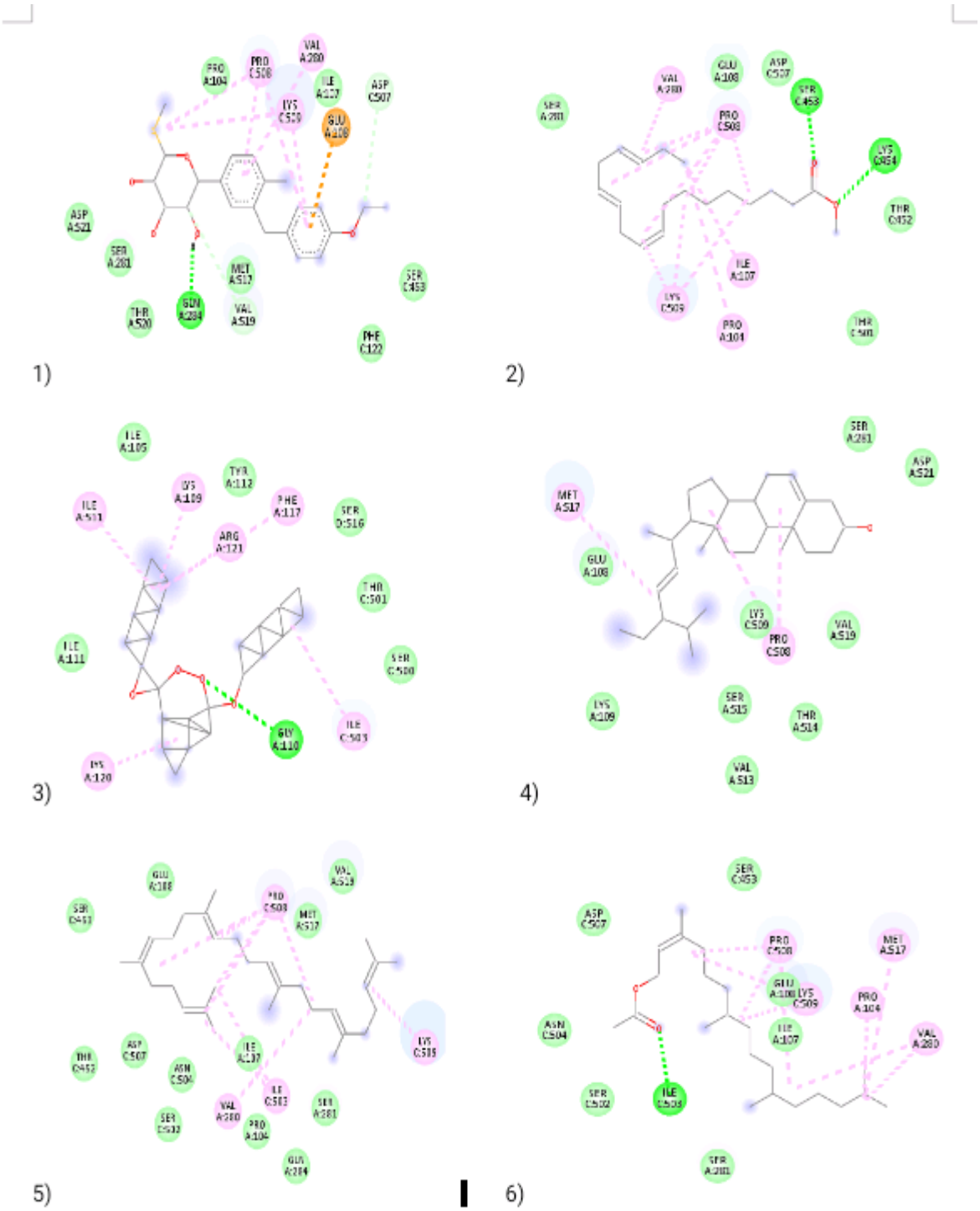


Fig 7: 2D representation of the interactions between the best pose found for (1) Sotagliflozin (Standard), (2) Glycerol-1-Palmitate, (3) Diisooctyl phthalate, (4) Stigmasterol, (5) Tetracosahexaene and (6) Phytol acetate with SGLT1 (3DH4).

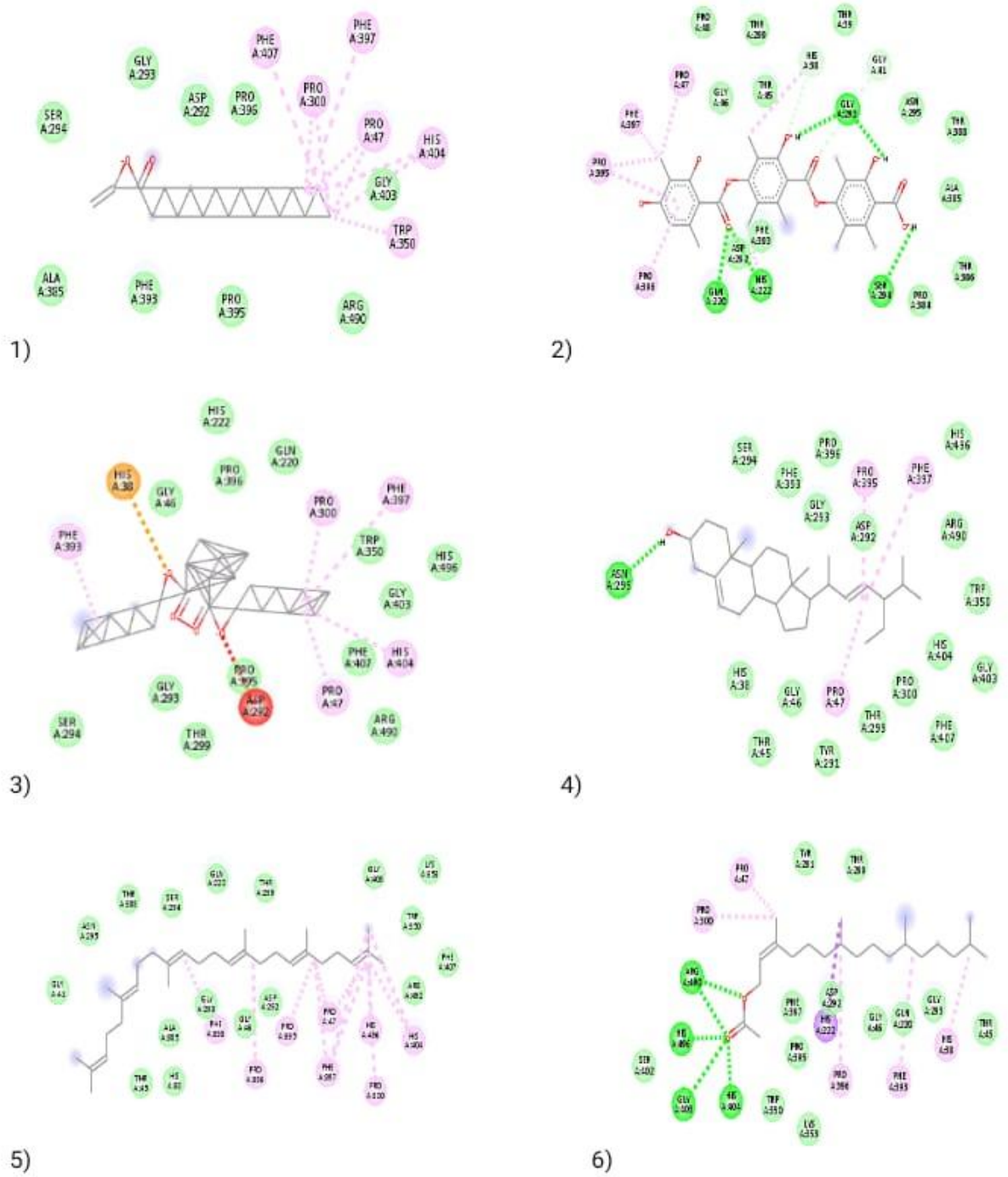


Fig 8: 2D representation of the interactions between the best pose found for (1) Acarbose (Standard), (2) Glycerol-1-Palmitate, (3) Diisooctyl phthalate, (4) Stigmasterol, (5) Tetracosahexaene and (6) Phytol acetate with α-GLU (1XSI).

Table 4: Protein-Ligands Interaction and Lig-Plot

SGLT1	Sotagliflozin (standard)	H-Bonding	GLN284(A), VAL519(A), ASP507(C)
		Hydrophobic interactions	PRO508(C), LYS509(C), VAL280(A).
	Glycerol-1-Palmitate	H-Bonding	SER453(C),LYS454(C),
		Hydrophobic interactions	PRO508(C), LYS509(C), VAL280(A), ILE107(A), PRO104(A).
	Diisooctyl phthalate	H-Bonding	GLY110(A)
		Hydrophobic interactions	ILE511(A), LYS109(A), ARG121(A), PHE117(A), ILE503(C), LYS120(A).
		H-Bonding	----------

	Stigmasterol	Hydrophobic interactions	MET517(A), PRO508(C).
	Tetracosahexaene	H-Bonding	----------
		Hydrophobic interactions	PRO508(C), LYS509(C), VAL280(A), ILE503(C).
	Phytol acetate	H-Bonding	ILE503(C)
		Hydrophobic interactions	PRO508(C), LYS509(C), VAL280(A), MET517(A), VAL280(A).
G6PC	(Standard)	H-Bonding	----------
		Hydrophobic interactions	PHE407(A), PRO300(A), PHE397(A), PRO47(A), HIS404(A), TRP350(A).
	Glycerol-1-Palmitate	H-Bonding	GLN220(A), HIS222(A), SER294(A), GLY293(A)
		Hydrophobic interactions	PHE397(A), PRO47(A), PRO395(A).
	Diisooctyl phthalate	H-Bonding	----------
		Hydrophobic interactions	PHE393(A), PRO300(A), PHE397(A), PRO47(A), HIS404(A).
	Stigmasterol	H-Bonding	ASN295(A)
		Hydrophobic interactions	PRO47(A), PRO395(A), PHE397(A).
	Tetracosahexaene	H-Bonding	----------
		Hydrophobic interactions	PRO300(A), PHE397(A), PRO47(A), HIS496(A), PRO47(A), PRO395(A), HIS404(A), PHE393(A),
	Phytol acetate	H-Bonding	ARG490(A), HIS496(A), GLY403(A), HIS404(A)
		Hydrophobic interactions	PRO300(A), PRO47(A), HIS222(A), PRO396(A), HIS38(A), PHE393(A),

Table 5 ADME/T properties Tetracosahexaene (6428538), Glycerol-1-Palmitate (14900), Stigmasterol (5280794), Diisooctyl phthalate (33934) and Phytol acetate (6428538)bySwissADME.

Pubchem ID of molecules	MW	HB donor	HB acceptor	LogP	Molar refractivity
14900	330.50	2	4	4.64	97.06
5280794	412.69	1	1	6.98	132.75
33934	390.56	0	4	6.50	116.30
6428538	338.57	0	2	6.78	108.68
6428538	410.72	0	0	9.38	143.48

a) Molecular weight (MW) (acceptable range: <500)
b) Hydrogen bond (HB) donor (acceptable range: ≤5)
c) Hydrogen bond (HB) acceptor (acceptable range: ≤10)
d) High lipophilicity (expressed as LogP, acceptable range: <5)
e) Molar refractivity should be 40-130.

IV. Conclusion

As Diabetes Mellitus is epidemic throughout the world, effective drugs with less/no toxicity need to be developed. And one such way is the use of herbal medicines which will have no side effects. Docking studies of bioactive compounds in *Pistiastrateotes* against two crucial drug targets for diabetes mellitus showed conducive results with prominent inhibitory activity was observed with Tetracosahexaene, Glycerol-1-Palmitate, Stigmasterol, Diisooctyl phthalate and Phytol acetate with inSodium/Glucose transporter-1 (SGLT1) and Glucose-6-phosphatase (G6PC) respectively. Thus the mentioned Phytochemicals (drugs) can be used for the developing a potent antidiabetic drugs.

Conflict the interest
The authors declare no conflict of interest

Authors' Declaration
The authors hereby declare that presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

References

[1]. WHO. (2016). Global Report on Diabetes. *Isbn*, *978*, 6–86.
[2]. American Diabetes Association. (2017). Examen de glucemia. *Diabetes Care*, *40*(Suppl 1), S11–S24. https://doi.org/10.2337/dc17-S005
[3]. Lotfy, M., Adeghate, J., Kalasz, H., Singh, J., & Adeghate, E. (2016). Chronic Complications of Diabetes Mellitus: A Mini Review. *Current Diabetes Reviews*, *13*(1), 3–10. https://doi.org/10.2174/1573399812666151016101622
[4]. Tulika, T., & Mala, A. (2014). Pharmaceutical Potential of Aquatic Plant Pistia stratiotes (L.) and Eichhornia crassipes. *Journal of Plant Sciences*, *3*(1), 10. https://doi.org/10.11648/j.jps.s.2015030101.12
[5]. Musa O (2005). Mineral composition of different parts of CapparisonilaDest Var. Cenescens (Coss) Heywood growing wild in Turkey.Journal of Medical Food 8 405-407.
[6]. Arber A (1991). The vegetative morphology of Pistiaand the Lemnaceae.Proceedings of the Royal Society 91 96- 103.
[7]. Khare CP. Encyclopedia of Indian medicinal plants. Berlin Heidelberg, Germany: Springer-Verlag. 2005. 372p.
[8]. Koffuor GA, Kyei1S,Woode E, EkuadziE, Ben IO. Possible mechanism of anti-inflammatory activity and safety profile of aqueous and ethanolic leaf extracts of PistiastratiotesL. J Ghana Sci Ass. 2012; 14(1):69-81.
[9]. Jha M, Ganesh N, Sharma V. In vitro evaluation of free radical scavenging activity of Pistiastratiotes. Asian Pac J Trop Dis. 2010; 2(1): 180-184.
[10]. Rahman MA, Haque E, Hasanuzzaman M, Muhuri SR, Shahid IZ. Evaluation of antinociceptive and antidiarrhoeal properties of Pistiastratiotes(Araceae) leaves.J PharmacolToxicol.2011; 6:596-601.
[11]. Lawal M, Suleiman A, Matazu NU, Dawud FA, Mohammed A, Umar IA. Antidiabetic Activity of *Pistiastrateotes*L. Aqueous Extract in Alloxan-induced Diabetic Rats. Trop J Nat Prod Res. 2019; 3(3):91-94.
[12]. International Journal of Basic and Applied Medical Sciences ISSN: 2277-2103 (Online) An Open Access, Online International Journal Available at http://www.cibtech.org/jms.htm 2017 Vol. 7 (1) January-April, pp. 14-21/Tyagi and Agarwal
[13]. Evans JL (2007) Antioxidants: do they have a role in the treatment of insulin resistance? Indian J Med Res 125(3):355
[14]. Gonzalez-Burgos E, Gomez-Serranillos MP (2012) Terpene compounds in nature: a review of their potential antioxidant activity. Curr Med Chem 19(31):5319–5341
[15]. Nguyen NDT, Le LT (2012) Targeted proteins for diabetes drug design. Adv Nat SciNanosciNanotechnol 3:013001
[16]. Deepak, K., Shirsat, R., & Kawale, M. (2018). an Overerview of Major Classes of Phytochemicals : Their Types an Overerview of Major Classes of Phytochemicals : Their Types and Role in Disease Prevention. *Hislopia Journal*, *9*(August), 1–11. https://www.researchgate.net/publication/327304552%0AAN
[17]. Madsen, P., & Westergaard, N. (2001). Glucose-6-phosphatase inhibitors for the treatment of Type 2 diabetes. *Expert Opinion on Therapeutic Patents*, *11*(9), 1429–1441.
[18]. Chothia C, LeskAM. The relation between the divergence of sequence and structure in proteins.EMBO J. 1986; 5:823-826.
[19]. Kaczanowski S, Zielenkiewicz P. Why similar protein sequences encode similar three-dimentional structures? TheorChem Acc. 2010; 125:643-650.
[20]. Read RJ, Adams PD, Arendall III WB, Brunger AT, Emsley P *et al*. A New generation of crystallographic validation tools for the protein data bank. Structure.2011; 19:1395-1412.
[21]. Savarino A. In Silico docking of HIV-1 integrase inhibitors reveals a novel drug type acting on an enzyme/DNA reaction intermediate. Retrovirol.2007; 4:1-21.
[22]. Rekha S, Chandrashekhara S. *Insilico*Proportional Molecular Docking Study and Analysis of Insulinotropic Activity of TZD Derivatives by PPARγ Activation. J. Pharm. Sci. & Res. 2017; 9(10):1799-1808.
[23]. Dennington R, Keith T, Millam J (2009) GaussView, version 5. Prod. Shawnee Mission Semichem Inc., Shawnee.
[24]. Dallakyan, S., Olson, A.J., 2015. Small-molecule library screening by docking with PyRx. In: Chemical Biology. Springer, pp. 243–250.
[25]. Daina, A., Michielin, O., Zoete, V., 2017. SwissADME: a free web tool to evaluate pharmacokinetics, drug-likeness and medicinal chemistry friendliness of small molecules. Sci. Rep. 7, 42717.
[26]. Devang, N., M, N., Rao, S., & Adhikari, P. (2016). HSD11B1 gene polymorphisms in type 2 diabetes and metabolic syndrome—Do we have evidence for the association? *International Journal of Diabetes in Developing Countries*, *36*(1), 95–102. https://doi.org/10.1007/s13410-015-0438-5
[27]. Ertl P, Rohde B, Selzer P. Fast calculation of molecular polar surface area as a sum of fragment-based contributions and its application to the prediction of drug transport properties, J Med. Chem. 2000; 43:3714-3717.
[28]. O'Boyle, N.M., Banck, M., James, C.A., Morley, C., Vandermeersch, T., Hutchison, G.R., 2011. Open Babel: an open chemical toolbox. J. Cheminf. 3 (1), 33.
[29]. Verma, A. K., Maurya, S. K., & Kumar, A. (2020). *Inhibition of multidrug resistance property of Candida albicans by natural compounds of parthenium hysterophorus L . An In- Silico approach*. *9*(3), 55–64.
[30]. Tyagi, T., & Agarwal, M. (2017). Gas chromatography-mass spectrometry analysis of bioactive constituents in the ethanolic extract of pistia stratiotes L. *International Journal of Basic and Applied Medical Sciences*, *7*(1), 14–21.
[31]. Trott, O., Olson, A.J., 2010. AutoDockVina: improving the speed and accuracy of docking with a new scoring function, efficient optimization, and multithreading. J. Comput. Chem. 31 (2), 455– 461.
[32]. Villa, F., Goebel, J., Rafiqi, F.H., Deak, M., Thastrup, J., Alessi, D. R., van Aalten, D.M., 2007. Structural insights into the recognition of substrates and activators by the OSR1 kinase. EMBO Rep. 8 (9), 839–845.
[33]. Sanner, M.F., 1999. Python: a programming language for software integration and development. J. Mol. Graph. Model. 17 (1), 57–61.
[34]. Wolber G, Langer T (2005) LigandScout: 3-D pharmacophores derived from protein-bound ligands and their use as virtual screening filters. J Chem