Original Article

COMPARISON OF COMMERCIALLY AVAILABLE DRUGS FOR TYPE 2 DIABETES WITH NATURAL MOLECULE FROM TINOSPORA

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ABSTRACT

Objective: Efficacy of natural molecule from *Tinospora cordifolia* versus commercially available drugs to control diabetes 2.

Methods: Twelve different drug molecules were selected to study drug properties, bioactivity and detailed mode of action. A comparative study was carried out among the drugs and plant metabolite to understand the putative mechanism of metabolite action and its potential to be developed as an herbal drug. PharmaGist Server was used to carry out pharmacophore modeling. The sequence of the target molecule (Q09428) was retrieved from UniProtKB/SwissProt, and structure prediction was carried out using ITASSER. The best model generated was further refined by energy minimization using Deep View. Validation of the structure was performed by Ramachandran plot analysis using PDBSum. Interaction analysis of the docked complex was done using LigPlot+.

Results: The potential of natural plant metabolite to target ATP-binding cassette sub-family C member 8 seems probable based on docking and interaction analysis results. The natural molecule showed comparable binding energy (-5.57) in four out of seven drugs.

Conclusion: Natural molecule from *Tinospora cordifolia* may serve as a potential lead drug molecule after modification and optimization for enhanced interaction.

Keywords: Diabetes mellitus, Natural molecule, Tinospora, Type 2 diabetes

INTRODUCTION

The type 2 diabetes is a chronic metabolic disorder triggered by insulin insensitivity and reduced level of insulin secretion. Insulin is a hormone that plays a key role in the transport of glucose to organs like the liver. Deficit production of insulin enhances the level of glucose in blood [1]. Type 2 diabetes, unlike type 1, is independent insulin disorder [2]. As per WHO 2014 factsheet, diabetes is prevalent in approximately 9% of adults. Though there is no cure for diabetes, drugs that stimulate insulin release are often used to regulate glucose absorption in type 2 diabetes patients. Sulphonylureas constitute a major group of insulin secreting anti-diabetic drugs [3]. Sulphonylureas function by targeting ATP-potassium channels in the membrane of pancreatic beta cells and inhibiting potassium efflux. Inhibition of efflux leads to depolarization and calcium influx. The influx enhances calcium-calmodulin binding, kinase activation, and release of insulin-containing granules by exocytosis. The drugs mimic the activity of glucose [4-7].

However, some findings associated use of sulphonylurea based drugs with hypoglycemia and increased risk of heart diseases [8]. Natural remedies and nutraceutical are hence preferred these days because; they provide the added advantage of minimum adverse effects. *Tinospora cordifolia* (Guduchi) is a medicinal plant that has been used since ancient times in the Ayurveda, for the treatment of multiple ailments including diabetes [9]. Secondary metabolites and active compounds of *Tinospora cordifolia* have been isolated from the plant and, the functions of these molecules have been reported. While both stem and root extracts of Tinospora regulate diabetes mellitus type 2; stem extract also functions in the regulation of diabetic neuropathy [10-13]. However, the detailed mode of action of active compounds has not been explored completely. This study focused on potential target identification for the insulin secretion stimulating active compound of *Tinospora cordifolia*. A comparative analysis has been carried out among the natural compound and some marketed sulphonylurea drugs to evaluate the applicability of Tinospora as a formulation for the development of an effective drug. Drugs used for the treatment of

diabetes mellitus type 2 were searched in drug bank and annotations of the records were compiled.

Twelve different drug molecules were selected to study drug properties, bioactivity (http://www. molinspiration. com) and detailed mode of action. Properties *viz.*, Drug Bank accession number, log P, polar surface area, the number of atoms, molecular weight, the number of polar groups with potential to form Hydrogen bonds, the number of violations, rotatable bonds, volume, and target of the drugs have been reported below (table1). Out of the twelve drugs studied, sulphonylurea class drugs function by blocking ATP-dependent K^+ efflux channel. The blockage leads to depolarization and activation of voltage-dependent calcium ion channel and Ca^{2+} influx, which eventually stimulates insulin release. A metabolite of *Tinospora cordifolia* has also been reported to stimulate insulin secretion [11] so; the comparative study was carried out among the drugs and plant metabolite to understand the putative mechanism of metabolite action and, potential to be developed as a drug. Properties of the drugs were analyzed, DB01251 has molecular weight 527.643Dalton which is not desirable according to Lipinski's rule of five [14].

Structures (http://www.molinspiration.com) of the sulphonylurea molecules were compared [fig.1] for pharmacophore identification; pharmacophore modeling was also carried out using the Pharma Gist Server [15]. Features of drug molecule reported by Pharma Gist were compared with plant metabolite. The sequence of the target molecule (Q09428) was retrieved from UniProtKB/SwissProt [16]. As the structure of target molecule has not yet been solved and, suitable templates with full-length query coverage were not identified, structure prediction was carried out using ITASSER [17]. In one run, ITASSER accepts query proteins are having less than 1500 residues so, structure prediction of the target was carried out in two separate jobs (position 1-1380 and 1290-1581) with an overlapping segment of 90 residues. Care was taken to ensure that regions are forming domains based on ProRule annotations [18] were intact as a query in one of the jobs. The best models generated by ITASSER were used as a template for Modeller9v14 [19] and, one intact model involving all the 1581 residues was generated. The best model generated was further refined by energy minimization using DeepView [20].

Validation of the structure was performed by Ramachandran plot analysis using PDBSum [21]. The refined model was used as the receptor for docking of the sulphonylurea drugs and natural ligand. Flexible docking was carried out using AutoDock4.0 suite [22], PyRx virtual screening software (http://pyrx. sourceforge. net/) was used to provide a graphical user interface for docking [23]. The structure predicted has distinct helix bundle supporting the confirmation of the transmembrane domain. The cavity-like structure associated with helix bundle was used to define the extracellular region by AutoGrid. LigPlot+ [24] was used for analysis of interactions in the docked complex; the plot confirmed the interaction of the ligand with residues in extracellular region proposed by Locate Database [25].

Table 1: Comparison of drug properties and targets for 12 approved drugs archived in drug bank and natural plant metabolite molecule

| Drug	logP	TPSA	natoms	MW	nON	nOHNH	nviolation	nrotb	volume	Target
DB01124	2.54	75.27	18	270.35	5	2	0	5	242.79	ATP-binding cassette sub-family C member 8
DB01382	1.19	90.42	21	309.38	7	1	0	7	259.11	ATP-binding cassette sub-family C member 8
DB01251	3.65	121.88	37	527.64	9	2	1	7	469.35	ATP-binding cassette sub-family C member 8
DB01120	1.45	78.5	22	323.42	6	2	0	3	284.59	ATP-binding cassette sub-family C member 8
DB00672	2.21	75.27	17	276.75	5	2	0	4	222.96	ATP-binding cassette sub-family C member 8
DB01016	4.77	113.6	33	494.01	8	3	0	8	424.74	ATP-binding cassette sub-family C member 8
DB01252	3.37	57.61	23	315.41	4	1	0	5	306.03	ATP-binding cassette sub-family C member 8
DB04876	1.42	76.36	22	303.41	5	2	0	3	289.82	Dipeptidyl peptidase 4
DB04878	-3.98	153.62	18	267.28	8	8	1	5	238.17	Dipeptidyl peptidase 4
DB06292	2.59	99.38	28	408.88	6	4	0	6	359.29	Sodium/glucose cotransporter 2
DB01132	3.07	68.29	25	356.45	5	1	0	7	318.53	Peroxisome proliferator-activated receptor gamma (Agonist)
DB08882	2.24	116.88	35	472.55	10	2	0	4	427.73	Dipeptidyl peptidase 4
Plant metabolite	-0.49	92.61	22	298.32	5	3	0	5	269.17	
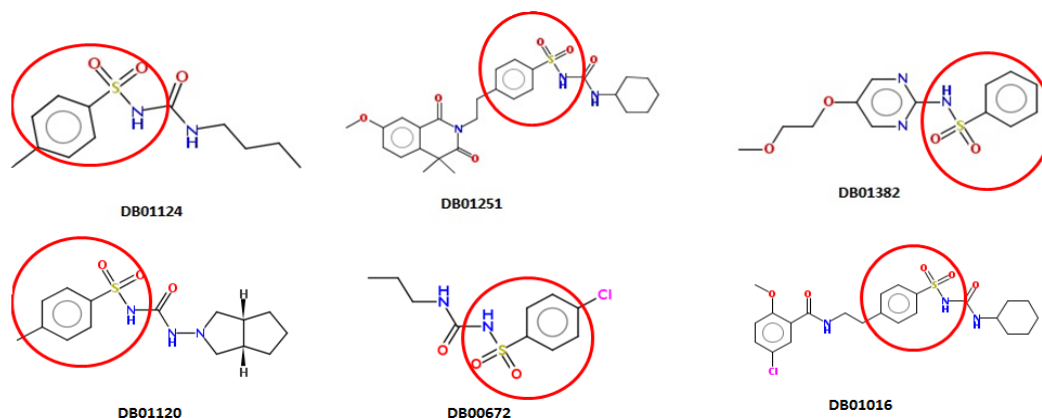
Fig. 1: Structure of six sulphonylurea drugs, the common unit has been encircled. Apart from the common aromatic ring, similar to peptide bonds, CO-NH groups are placed in trans conformation in five drugs
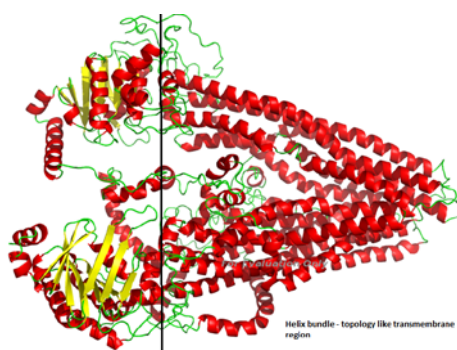

Fig. 2: Predicted structure of target protein represented in cartoons form, distinct helix bundle structure expected to form the transmembrane region is observed. Helix regions shown in red colour constitute 56.2% region of the structure, yellow colored strands form 7.6%

Extracts of the plant *Tinospora cordifolia* have been used for the treatment of many diseases since ancient times. Researchers have identified multiple active compounds of the plant, one of which has been reported to possess insulin release stimulating effect. While activity and compound details have been reported, details of the mode of action of the plant metabolite have not been reported [11]. The current work focused on the comparison of the natural molecule with approved drugs having similar insulin release stimulating effect. Sulphonylurea drugs target ATP-binding cassette sub-family C member 8 and stimulate insulin release. The potential of natural plant metabolite to target ATP-binding cassette sub-family C member 8 seems probable based on docking and interaction analysis results. The optimum model [fig. 2] of target generated after energy minimization had only 1.3% of the non-proline, non-glycine residues in the disallowed region of Ramachandran plot whereas, 84.4% residues were found in the most favored regions. PDBSum reported a total of 11 pores which collectively span through the predicted structure and thus justify the function of transport across the membrane. The model predicted also has helix bundle structure like most transmembrane protein receptors.

Table 2: Summarizes the details of binding affinity and drug-target interaction. Binding energy of the natural molecule is comparable with chemical drugs

	Binding energy	Interacting residues
DB01124	-5.67	Hydrophobic interaction–L508, L511, Y512, R1418, I1423, L1425, D1427, V1429. Polar contact-R1497, H-bond at 2.84Å.
DB01251	-6.17	Hydrophobic interaction–K507, L508, L511, Y512, S1385, L 1389, F1392, I1423, L1425, Q1426, D1427, V1429. Polar contact-D1505, H-bond at 3.12Å and 2.6Å.
DB01382	-4.02	Hydrophobic interaction–I60, H61, T64, H67, E88, W514, F1431, S1432, F1437, P1441, D1442.
DB01252	-6.27	Hydrophobic interaction–L511, Y512, W514, R1418, S1419, P1441, K1444, R1497.
DB00672	-5.09	Hydrophobic interaction–L511, Y512, R1418, S1419, R1493, R1497.
DB01016	-3.64	Hydrophobic interaction–W1338, I1456, T1525, A1526, A1528, D1529, R1530.
DB01120	-4.21	Hydrophobic interaction–N1365, A1366, L1367, L1547, I1556, F1559.
Natural ligand	-5.57	Hydrophobic interaction–L508, L511, Y512, I1423, L1425, D1427, V1429, R1497.

Though the natural metabolite does not show the best affinity, it is comparable with the chemical drugs group. Natural molecules are usually preferred as treatment measure because of their minimum adverse effects. *Tinospora cordifolia* is a plant with multiple medicinal benefits, and it has also been reported to stimulate insulin production. The present study focused on the comparison of active compound of Tinospora with drugs having insulin production stimulating function. Though it has not shown the best interaction with the target protein, however, the binding energy and interactions are comparable. This plant metabolite may thus serve as a potential lead molecule for further derivatization, optimization, and enhanced interaction.

CONFLICT OF INTERESTS

Declared none

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