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Research Article

STRUCTURE BASED DRUG DESIGN STUDIES ON HETEROARYL PROPANOIC ACID DERIVATIVES AS PPARΓ AGONISTS

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ABSTRACT

Peroxisome proliferator-activated receptors (PPARs) are a group of nuclear receptor proteins. Docking studies are based on several factors. Among 15 entries of PPARγ, 2Q6S was taken for docking analysis, as it showed 418 most favored regions, 35 in additionally allowed region and none of the residue in disallowed regions. To carry out drug designing, molecules were considered from the literature in which substitution of R_1 position with dihydrofuryl reported to have high dock score (-14.98 Kcal/mol) than the remaining analogues, with better geometry and interactions. Hence docking analysis using heteroaryl propionic acid derivatives as anti-diabetic agents suggest the reproducibility of active molecules being predicted by computational docking studies using Auto dock software.

KEY WORDS: PPAR gamma, propionic acid, Docking, AutoDock

INTRODUCTION:

Research Article

Diabetes mellitus (Type II) is a metabolic disorder which is characterized by dysfunctioning of pancreatic beta cells along with insulin resistance, if not controlled leads to macro and microvascular disorders[1, 2]. Dipeptidyl peptidase IV[3], GLP-1 analog[4], Glucokinase[5], PPAR (peroxisome proliferated activated receptor) [6] have been identified as potential targets of type II diabetes. Peroxisome proliferator-activated receptors (PPARs) are a group of nuclear receptor proteins. They play essential role in the cellular metabolism of carbohydrates, lipids, and proteins, cell differentiation and development[7, 8]. PPARs function as transcription factors[9] regulating the expression of genes[10]. These are of three types i.e. alpha (α), delta (beta) (β/δ), gamma (γ)[8]. The molecular target of the glitazones was reported to be PPAR-gamma[11] which is expressed in three forms; they are gamma-1 (γ1), gamma-2(γ2), gamma-3(γ3). The role of PPAR in combating diabetes[12] has provided us the rationale to carry out structure based drug design studies[13].

The selection of protein for docking studies is based upon several factors like, it should contain a ligand, structure should be determined by X-ray diffraction, and resolution between 2.0-2.5 Angstroms and out of the 15 entries of PPARγ, 2Q6S was taken for docking analysis (based on the Ramachandran plot statistics) as it showed 418 most favored regions, 35 in additionally allowed region and none of the residue in disallowed regions. ISIS draw 2.6, AutoDock 3.0 (an automated docking tool) and Web Lab Viewer 4.0 were used as computational tools. A graphical user interface called Auto dock Tools, or ADT was utilized to generate grids, calculate dock score and evaluate the conformers.

COMPUTATIONAL TOOLS

Structural based drug designing:

Structure-based drug design is one of several methods in the rational drug design toolbox. Drug targets are typically key molecules involved in a specific metabolic or cell signaling pathway that are known, or believed, to be related to a particular disease state. Drug targets are most often proteins and enzymes in these pathways. Molecules are designed to inhibit, restore or

otherwise modify the structure and behavior of disease-related proteins and enzymes. Structure-based drug design uses the known 3D geometrical shape or structure of proteins to assist in the development of new drug molecules.

AutoDock:

AutoDock is an automated docking tool. It is designed to predict how small molecules, such as substrates, bind to a receptor of known 3D structures. AutoDock actually consists of two main programs: one performs the docking of the ligand to a set of grids describing the target protein; and the other Auto Grid pre-calculates these grids. In addition to using them for docking, the atomic affinity grids can be visualized. A graphical user interface called AutoDock Tools or ADT was utilized to generate grids, calculate dock score and evaluate the conformers.

WeblabViewerlite:

The WebLab Viewer is an innovative software tool for examining the 3D structure of molecular models, and for communicating the resulted information. With the WebLab Viewer, a molecule can be viewed as a wireframe, a stick model, a ball and stick model, or a space-filling model. The model can be rotated, translated, or scaled to any particular viewpoint. Distances, angles, torsions, and stereochemistry can be easily measured; these variables are instantly updated whenever the local geometry is modified.

MATERIALS AND METHODS

A total of 15 entries of PPARγ were selected from RCSB protein data bank, based on the presence of ligand, x-ray diffraction and 2.0-2.5 Aº resolution. Out of the 15 entries, 2Q6S was taken for docking analysis (based on the Ramachandran plot statistics) as it showed 418 most favored regions, 35 in additionally allowed region and none of the residue in disallowed regions. A comparative protein - ligand dock analysis was performed using 2Q6S extracted from Protein Data Bank (PDB) [14] to evaluate the algorithm and scoring function efficiency between AutoDock

3.0 and experimental activities. The molecules selected for docking studies are from the selected article[15, 16].

All these molecules as well as the bound ligand of the protein 2Q6S were docked by using the software AutoDock and the score values are predicted. The protein ligand interactions were also studied in web server. Based on the score values against the activity in μM the molecules were represented as active, moderately active and inactive. All molecules were drawn using Integrated Scientific Information System (ISIS) draw tool and energy minimized using Tsar Software. Automated docking was used to locate the appropriate binding orientations and conformations of various inhibitors into the 2Q6S binding pocket. To perform the task, the powerful genetic algorithm method implemented in the program AutoDock 3.0[17] was employed. All water molecules were removed from the original Protein Data Bank file. Polar hydrogen atoms and Kollman charges[18] were added. Grid maps were generated by AutoGrid program. Each grid was centered at the crystal structure of the corresponding 2Q6S bound ligand PLB5001 (B). The grid dimensions were 60 A˚ X 60 A˚ X 60 A˚ with points separated by 0.375 A˚. For all ligands, random starting positions, random orientations and torsions were used. During docking, grid parameters were specified for x, y and z axes as 38.808, 30.946 and 42.249 respectively.

RESULTS AND DISCUSSION

Molecules selected from the articles[15, 16] were docked using AutoDock software and docked scores of those molecules were represented in Table-1, with their activity, number of hydrogen bonds and interacting residues. Molecules with high dock scores were selected for regression analysis and their docked scores along with their activity, number of hydrogen bonds and interacting residues were given in Table-2. The newly designed molecules in Table-3 are energy minimized and the resulting molecules are considered for docking analysis using AutoDock 3.0 in Linux. Ligand and proteins were prepared. At the end of each run, docked

orientations are saved and the resultant molecules are checked for geometry and no of hydrogen bonds using web lab viewer software. AutoDock is employed to study the docking molecules within active site region of 2Q6S and web lab viewer is used to study the H-bond interaction. Docked scores of newly designed molecules are represented in Table-4.

The newly designed molecules (MOL_NW) were docked against the protein 2Q6S and the dock scores were reported in Table-4 above and the following interactions were seen with the new ligands and the active sites of the protein. From Table-4, it became evident that the newly designed molecules have docked scores more than -12.71kcal/mol, which is the docked score of 2Q6S. Hence, further proof was provided by plotting a graph (Figure-1) between experimental values and dock scores, where it is clear that they represented a correlation of 0.7489.

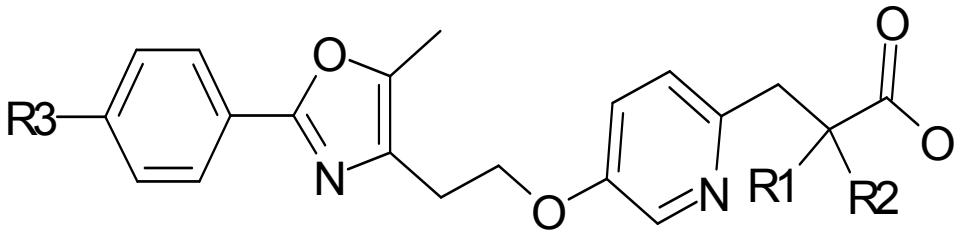
Table 1: The ligands selected for docking studies and their corresponding interaction energies.

Molecules	Activity (K_i) in µM	AutoDock score (Kcal/mol)	No. of H-bonds	Interacting residues
1	2.1	-9.16	2	Glu259, Ile281
2	1.1	-13.12	2	Glu559, Ile281
3	0.24	-13.53	1	Lys265
4	8.8	-12.13	1	Phe265
5	0.36	-12.4	1	Lys265
6	12.0	-11.93	2	Ser342, Lys265
7	5.1	-7.73	1	Arg288
8	0.063	-13.04	1	Lys265
9	0.15	-13.09	1	Arg288
10	0.56	-10.83	1	Lys265
11	14.4	-11.19	1	Glu291
12	7.98	-12.92	1	Lys265
13	0.394	-12.88	1	Lys265
14	3.5	-12.91	3	Arg288, Cys285, Ser289
15	0.038	-9.44	0	-
16	17.0	-13.52	1	Lys265
17	0.081	-13.67	1	Ser289
18	0.81	-13.05	2	Ile281, Cys285
19	0.023	-13.42	1	Arg288
20	0.016	-14.65	1	Ile281
21	0.052	-12.94	2	Lys367, Cys285
22	0.011	-13.12	1	Ser342
23	0.094	-13.74	0	-
24	0.015	-13.72	0	-
25	0.017	-13.05	1	Arg288
26	0.007	-14.6	1	Arg288
27	0.334	-12.35	3	Ile281, Cys285, Ser289
28	0.27	-10.9	0	-
29	0.006	-13.78	2	Gly284, Lys265
30	0.348	-11.46	1	Ile326
31	0.073	-12.45	1	Met348
32	1.6	-12.95	2	Lys265, His266
33	0.21	-13.61	1	Arg288
34	1.1	-13.12	0	-
35	1.1	-11.34	2	His266, Ile281
36	0.093	-12.59	4	Ile281, Gly284, His266, Lys265
37	0.3	-13.63	2	Gly284, Lys265
38	0.02	-13.27	2	Ser289, Cys285
39	0.043	-12.04	1	Ile281
40	21.0	-14.09	1	Ile281
41	0.043	-13.21	2	Arg288, Leu340

Table 2: Dock scores of molecules showing experimental activity and computational binding energy values for set of molecules under study.

Molecules	Activity (K_i) in µM	AutoDock score (Kcal/mol)	No. of H-bonds	Interacting residues
8	0.063	-13.04	1	Lys265
19	0.023	-13.42	1	Arg288
21	0.052	-12.94	2	Lys367, Cys285
22	0.011	-13.12	1	Ser342
24	0.015	-13.72	0	-
29	0.006	-13.78	2	Gly284, Lys265
31	0.073	-12.45	1	Met348
36	0.093	-12.59	4	Ile281, Gly284, His266, Lys265
38	0.02	-13.27	2	Ser289, Cys285
41	0.043	-13.71	2	Arg288, Leu340

Table 3: Newly designed molecules from the basic structure.

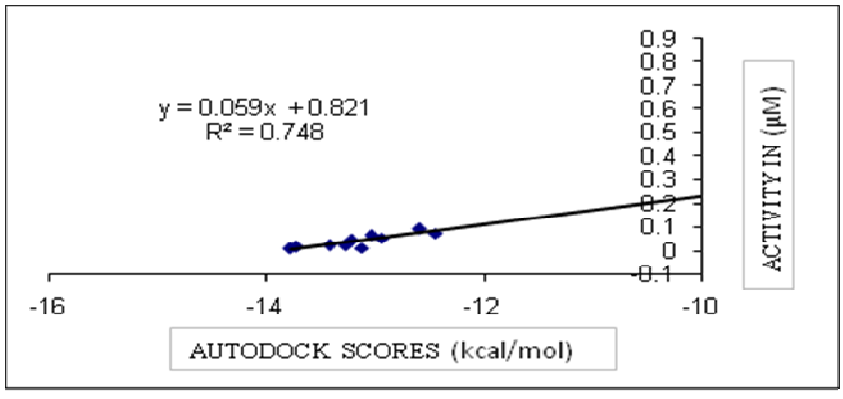


S.No	R1	R2	R3
1	propyl	H	propyl
2	dihydrofuryl	H	propyl
3	sulphur	H	propyl
4	chlorine	H	propyl
5	propyl	H	propyl
6	Isopropyl	H	propyl
7	bromine	H	propyl
8	isobutyl	H	propyl
9	H	propyl	propyl
10	H	sulphur	propyl
11	H	chlorine	propyl

Table 4: Docked scores of newly designed molecules (MOL_NW).

S.No of MOL_NW	AutoDock score (Kcal/mol)	No. of H-bonds	Interacting residues
1	-13.62	1	Ile326
2	-14.98	2	Glu343 Arg288
3	-13.01	3	Glu284 Ile281 Cys285
4	-13.83	2	Lys265 Gly284
5	-14.05	1	Ser289
6	-13.83	2	Gly284 Ser342
7	-14.0	0	-
8	-14.92	1	Lys265
9	-14.43	0	-
10	-13.71	4	His266 Gly284 Arg280 Ile281
11	-13.57	1	His266

Figure 1: Graph plotted between dock scores and activity values of selected molecules from Table-2.



CONCLUSION

The RMSD (Root Mean Square Division) observed was reported to be less than 2 Aº with respect to 2Q6S bound ligand, justifies the validation of AutoDock software. Inhibitors exhibited positive correlation with experimental data where in highly active molecules showed high dock score (MOL_NW-2, -14.98 & MOL_NW-8, -14.92) and similar observation was also reported in moderately active molecules (MOL_NW-4, -13.83 & MOL_NW-5,-14.05) and least active (MOL_NW-3, -13.01 & MOL_NW-11, -13.57)**.** Gly284, His266, Lys265 H-bond interactions are represented in most cases. The docking program in auto dock justifies the correlation between the experimental values and the values derived computationally. In these newly designed molecules with novel analogues, MOL_NW-2 reported to have high dock score (-14.98 Kcal/mol) than the remaining analogues. In the 2^{nd} molecule R_1 position was substituted by dihydrofuryl to enhance the features of the designed molecule. Therefore, the dock analysis performed in AutoDock suggests the importance of evaluating the prediction accuracy of scoring functions adopted in various docking programs. In AutoDock, a positive correlation was observed between experimental values and computational dock scores.

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