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Docking studies of Prasugrel by using MolDock software

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Abstract: Prasugrel is the member of the thienopyridine class of ADP receptor inhibitors. This agent reduces the aggregation (clumping) of platelets by irreversibly binding to P2Y12 receptors. The structure of Prasugrel was drawn using Chemsketch software and docking studies of this compound have been performed using MolDock software. The result shows that Prasugrel is involved in the hydrogen bond formation with Thr 302 in the active site and the best conformation of this compound is obtained in which it is more active as P2Y12 receptor antagonist and reduces platelets aggregation more effectively.

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INTRODUCTION

Platelet plays essential roles in hemostasis and arterial thrombosis[1]. After thromthrombogenic stimuli such as vessels injury or lesions rapture in a coronary artery, platelets are activated by adhesion to the damaged sites, which further initiates a cascade of downstream signaling and ultimately leads to the occurrence of adverse cardiovascular events[2, 3]. During this complicated process, a number of mediators released from the platelets and the receptors on platelet membrane are involved, among which adenosine triphosphate (ADP) and its corresponding platelet receptors are great importance[4]. ADP, mainly secreted by erythrocytes, endothelial cells and dense granules of stimulated platelets, serve as a powerful agonist in the platelets aggregation[5]. Effects of ADP is manifested through two distinct platelets receptors, namely P2Y1 and P2Y12, both of which are purinoreceptors belonging to the G-protein coupled receptor family[6]. Due to unique role in platelets activation and aggregation, the blockage of interaction between ADP and its receptors is of great value forselective attenuation of ADP-induced platelet activation. Compared with the ubiquitously expressed P2Y1, P2Y12 is mainly found on the surface of human platelets, making it a more attractive target for the development of novel anti-platelet agent[7].

Based on chemical properties and inhibitory mechanism P2Y12 receptors are divided in two types: Thienopyridines including Ticlopidine, Clopidogrel and Prasugrel have been more commonly used in clinical practice nowadays[8]. In general, the thienopyridine drugs share similar inhibitory mechanism. They all are prodrugs that have to be converted first to the activated forms via hepatic cytochrome P450. The activated metabolite can specifically and irreversibly bind to cysteine residues of the P2Y12 receptor and consequently inhibit the ADP mediated platelet activation and aggregation[9].

Prasugrel chemically is 5-[2-cyclopropyl-f great valu1-[2-fluro-phenyl]-2-oxoethyl]-4, 5, 6, 7-tetra hydrothieno [3, 2-C] pyridine-2-yl acetate. It is the member of the thienopyridine class of ADP aggregation (Clumping) of plateletsby irreversibly binding to P2Y12 receptors. Prasugrel inhibits adenosine diphosphate-induced platelet aggregation more rapidly, more consistently, and to a great extent than do standard and higher doses of Clopidogrel in healthy volunteers and in patient with coronary heart diseases[10,11].

Prasugrel is specifically indicated to reduce the rate of thrombosis in patient with ACS who is to manage by PCI, including patients with unstable Angina or non-ST -elevation MI when manage with primary or delayed PCI. The effectiveness of new drug was demonstrated in studies in which it was used in conjunction with Aspirin and compared with regimen of Clopidogrel plus Aspirin. The primary outcome measure was the composition of cardiovascular death, non fetal MI and non fetal stroke. When compared with the Clopidogrel/Aspirin regimen, the Prasugrel/Aspirin regimen provided a 19% relative risk reduction in non fetal MI.Fewer stent-related clots (i.e. stent thrombosis) were also observed in patient treated with Prasugrel/Aspirin with a relative risk reduction approximately 50%[12].

Molecular docking is a simulation process to predict the conformation of a receptor-ligand complex, where the receptor can be a protein and the ligand a small molecule. It can also be defined as a simulation process where a ligand position is estimated in a predicted or pre-defined site in the receptor molecule. All structural based virtual screening projects are based on the hypothesis that it is computationally determining the three-

dimensional structure of binary complexes involving a protein and a ligand.

Here, in this study Molgro Virtual Docker (MVD) software is used for docking studies. It is based on a new hybrid search algorithm, called Guided differential evolution. Differential evolution was introduced by Storn and Price[13] in 1995 and has been applied previously for docking purpose[14]. The docking scoring function of MVD is based on a piecewise linear potential (PLP) introduced by Gehlhaar et al.[15,16]and further extended in GEMDOCK by Yang et al[17]. The scoring function was further improved to include new hydrogen bonding term and new charge schemes[18].

This MVD software has shown a higher docking accuracy as compare to the other available docking software (Table 1) with respect to the identification of ligand-binding modes[19].

Table 1: Comparison of docking accuracy and average RMSD values of MolDock with other available docking programs[17].

Method	Docking Accuracy	Average RMSD (Å)
MolDock	87.01%	1.38
Glide	81.82%	1.38
Surflex	75.32%	1.86
FlexX	57.89%	3.15

MATERIALS AND METHODS

The structure of Prasugrel was drawn by using CHEMSKETCH software. The three dimensional structure of human CYP 450 2B6 was retrieved by Protein Data Bank (http://www.rcsb/pdb; accession codes 3IBD). For docking purpose, MVD software is used in this study because of its higher accuracy. This software automatically identifies the potential binding site by using the cavity detection algorithm which also includes the bound ligand (Figure 2).

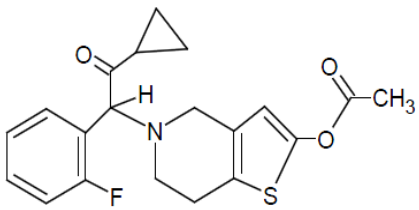

Figure 1: Structure of Prasugrel.

Ten independent docking runs were performed here in this study and each of these docking run come up with one solution (pose). Thus 10 solutions (poses) obtained from 10 independent docking runs (Figure 3). Each pose was inspected individually and the interaction of the docked compound with the amino acids in the binding site is viewed. The pose selected as the best is the one with the highest MVD score.

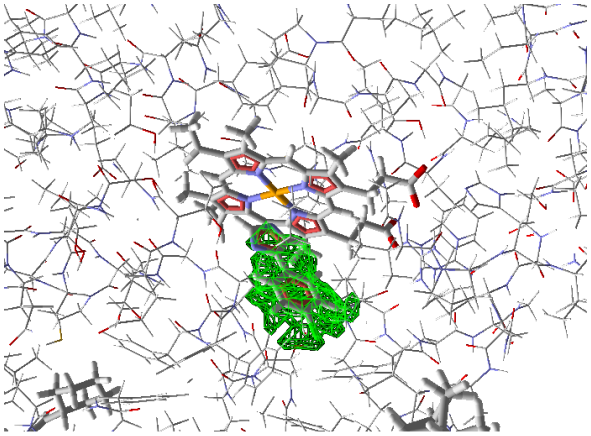

Figure 2: The binding cavity (in green color) identified by by MVD.

RESULTS AND DISCUSSION

The crystal structure of CYP450-2B6 (PDB ID: 3IBD) enzyme is used here for docking studies. In this crystal structure the enzyme is bound to a ligand 4-(4-chlorophenyl)imidazole (CPZ), which forms hydrogen bond interactions with the amino acid residues Thr302 within the active site of this enzyme (Figure 4). Interactions of the ligand with this amino acids have been shown to be crucial for the activity of this enzyme[20]. Prasugrel is docked with in the active site of CYP 450 2B6 (PDB ID: 3IBD) and it come up with 10 poses. Table 2 presents the MVD score for all these 10 poses obtained upon docking of Prasugrel and the best pose with the highest score is selected (Figure 5).

Table 2: MVD score for 10 poses obtained upon docking of Prasugrel.

Ligand	MVD Score
Pose1	-89.854
Pose2	-70.9677
Pose3	-56.9181
Pose4	-52.5547
Pose5	-45.6651
Pose6	-42.2731
Pose7	-33.7688
Pose8	-11.8828
Pose9	-8.13943
Pose10	173.294

The docking result shows the Prasugrel conformation (with lowest energy) in which it will be most active and has formed the hydrogen bonds

with the active site residues Thr302 and Ileu101 (Figure 5).

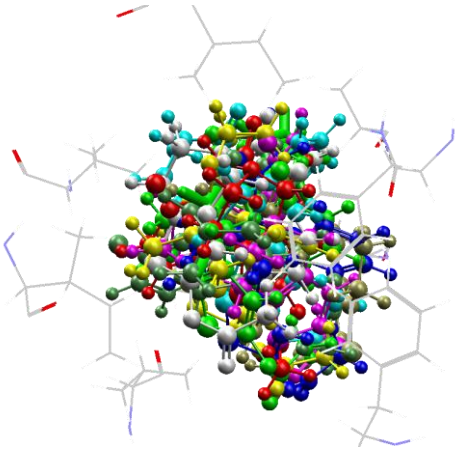

Figure 3: Ten poses obtained after docking of Prasugrel. Each pose is presented with a different color.

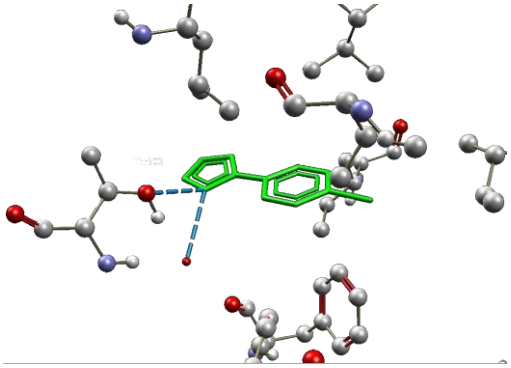

Figure 4: The binding of 4-(4-chlorophenyl)imidazole with in the active site of CYP 450 2B6. (The amino acids in the active site are presented in ball and stick with element colour and the bound ligand is presented with green colour. Blue lines represent the hydrogen bonds in between the ligand and the active site).

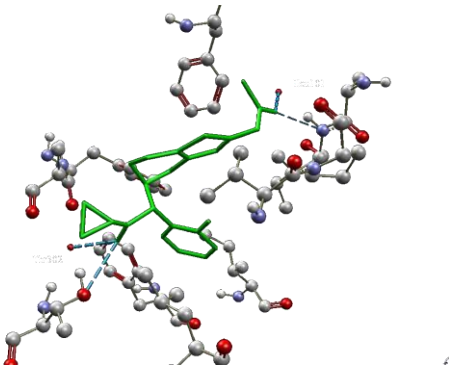

Figure 5: The best scored docking solution of Prasugrel with the CYP 450 2B6. (The amino acids in the active site are presented in ball and stick with element colour and the docked Prasugrel is presented with green colour. Blue lines represent the hydrogen bonds in between the ligand and the active site).

Comparison of figures 4 and 5 shows a similar mode of binding of the docked Prasugrel, and the bound ligand 4-(4-chlorophenyl)imidazole in the active site of the crystal structure of CYP 450 2B6, both the two are forming hydrogen bonds with active site residue Thr302, thus, Prasugrel in this conformation could be a potent inhibitor of ADP receptor of class thienopyridine.

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