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Original Article

AFFINITY OF WARFARIN WITH CYP2C9 BY MOLECULAR DOCKING STUDY

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ABSTRACT

Objective: Ligand-protein docking has been introduced as a computer method for identification of potential protein targets of a drug. A protein structure database is searched to find proteins to which a drug can bind or weakly bind information from protein data bank this study is designed.

Methods: With the aid of the automatic molecular docking, the affinity of CYP2C9,for warfarin was studied by using Pymol and Auto dock-Vina software programs

Results: Molecular Docking Results showed that CYP2C9 interaction energy & bond distance (-10.2) (3.0, 3.6) with Warfarin, The most important residues for enzyme–substrate complexes, such as Phe100, Ala 103 of CYP2C9 were identified.

Conclusion: From this study we suggest CYP2C9–Warfarin complex has higher stability and stronger affinity because CYP2C9 shows more favorable interaction energy.

Keywords: Cyp2c9**,** Warfarin, Molecular Docking, Pymol, Autodock-vina.

INTRODUCTION

Cytochrome P450 (CYP) 2C9 is the principal isoform of the CYP2C subfamily in the human liver and is involved in the oxidation of several endogenous and xenobiotic compounds, which plays major role in Metabolism of warfarin.Warfarin is Coumarin derivative act as an Oral Anticoagulant,its Molecular Formula: $C_{19}H_{16}O_4$& its Systematic name: 4-Hydroxy-3-(3-oxo-1-phenylbutyl)-2H-chromen-2-one, its Anticoagulant effect bycyclic conversion of vitamin K and vitamin K epoxide.3months anticoagulants therapy is required in VTE it includes- Deep Venous Thrombosis (DVT), Pulmonary Embolism (PE)& also used in treatment of Rheumatic heart disease. The metabolism of warfarin by CYP2C9 can yield either safe or toxic products, which may be related to the recognition and binding modes of the substrates to this isoform. These interactions can be studied by using molecular docking studies. Therefore, the current study emphasizes the interaction energy of warfarin with CYP 2C9 and the important residues for enzyme substrate complexes are known by using computational methods for ligand- protein interaction.

Observations

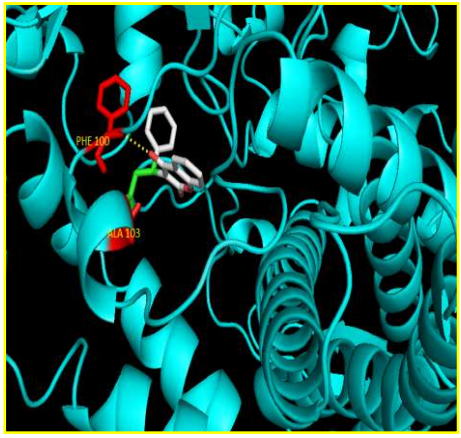

Fig. 1: it shows amino acid residues of cyp2c9 interaction with warfarin (cartoon model)

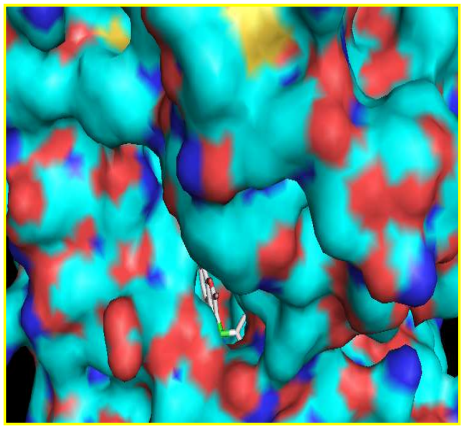

Fig. 2: it shows surface model of cyp2c9- warfarin interaction.

MATERIALS AND METHODS

With the aid of the automatic molecular docking, the affinity of CYP2C9, for warfarin, was studied by using Pymol and Auto dock-Vina software programs.

PDB Search

Protein& ligand informationretrieved from the Protein Data Bank& Drug bank to study the Interaction of warfarin with CYP2C9. It is given pdb id is 1OG5 [1] depending on chain involvement with warfarin, the ptn.pdb file had been prepared.

MGL tool & PDBSUM

MGL tool & Pdb sum were used to prepare grid to check whether residues inside or not and then pdb.qt and ligand pdb.qt files prepared& then to get configuration, command is given by using (auto dock) vina.exe [2] which had given ligand out pdbqt.

Visualization & labeling

After the formation of all files protein& ligand both were visualized by using pymol. From the docked complex interacting residues were

identified and labeled them. From this methodology we report the interacting residues with their interacting energy for enzyme substrate complex, and also bond distance is known by finding the polar contacts with the method of surface representation Docking technique.

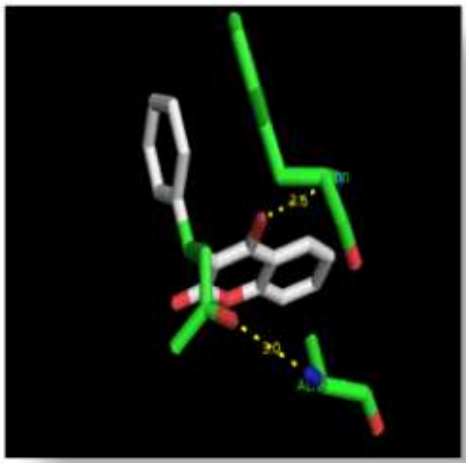

Fig. 3: it shows bond distance in warfarin-cyp2c9 interaction

RESULTS

Affinity of warfarin with CYP2C9 enzyme is -10.2(kcal/mol) Interaction energy, and the Important residues for enzyme substrate complex identified as Phe 100, Ala 103& the Bond distance between interacting residues and warfarin is (3.0, 3.6) These results may provide a possible molecular basis for understanding complex drug-drug & drug-protein interactions.

DISCUSSION

Warfarin and homologous compounds derived from coumarin are the most widely used oral anticoagulant drugs worldwide. They function as antagonists of vitamin K, an essential cofactor for the posttranslational γ-glutamyl carboxylation so called as vitamin K-dependent proteins. [3] The CYP2C9 ranks amongst the most important drug metabolizing enzymes in humans, S-warfarin is Substrate for CYP2C9,Approximately 80 to 85% of the more potent (S)-enantiomer of warfarin is eliminated by CYP catalyzed biotransformation to 6- and 7-hydroxy (S)-warfarin, CYP2C9 functions as catalyst for the 6- and 7-hydroxylation of (S)-warfarin at therapeutically relevant substrate concentrations. [4] Polymorphisms in the coding region of the *CYP2C9* gene produce variants at amino acid residues 144 (Arg144Cys) and 359 (Ile359Leu) of the CYP2C9 protein. Individuals homozygous for Leu359 have markedly diminished metabolic capacities for most CYP2C9 substrates, although the frequency of this allele is relatively low.

Here we report that in humans warfarin shows most probable interaction energy at CYP2C9 and the important amino acid residues involved at the active site are noticed as phe100, ala 103.These residues may be involving in the metabolism of warfarin. Mutation in this CYP2C9 gene causes configurational changes in enzyme CYP2C9 and the variants at amino acid residues involved in the enzyme substrate complexes will be worked out following this work to predict potential toxicity and side effect of anticoagulants.

CONCLUSION

CYP2C9–Warfarin complex has higher stability and stronger affinity with more favorable interaction energy. By analyzing the results we suggest that warfarin showing direct interaction with CYP2C9 which is a major enzyme in its metabolism.

FUTURE WORK

By applying these molecular modeling techniques: (1) identification of unknown and secondary therapeutic targets of Anticoagulants, (2) those will help in drug discovery.

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