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Molecular Docking Analysis of *Chlamydia pneumonia* Derived Protein

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Abstract—*Coronary Artery Disease (CAD) is a leading cause of mortality and morbidity worldwide, due to asymmetric focal thickenings of the intima. They consist of cells, connective-tissue elements, lipids, and debris. Chlamydia pneumoniae is a gram-negative bacterium associated with respiratory tract infection of humans and it is also assumed to play significant role in Atherosclerosis patients. Studies revealed the significant role of C.pneumonia in the procured inflammation and are the only viable pathogen present in the atherosclerotic plaques. This pathogen plays its crucial role in the phenomenon of oxidation of lipoproteins and the induction of cytokines and proteolytic enzyme production, which are essential for Atherosclerosis. Three virulent, membrane proteins of C. pneumoniae were predicted to propose them as candidate proteins for vaccine production against chlamydial infection. The identified RVOM1 protein is a translocator protein involving the auto-transporter domain and the missing N-terminal passenger domain. Auto-transporters are synthesized with an N-terminal signal peptide to convey transportation across the inner membrane through the secretary machinery. Effect of anti-inflammatory compound from medicinally valued plants revealed Stigmasterol as a potential inhibitor. In the present study docking of stigmasterol with RVOM1 was carried out with Autodock 4.2. Lead characterization was done by Molinspiration and Lazar toxicity prediction server. Binding studies shows binding energy -6.45 kcal/mol and inhibitory constant 18.66 µm. Result indicates that it could be possibly used in drug designing studies to minimize the effect of inflammation.*

1. INTRODUCTION

Coronary artery disease (CAD) is an inflammatory disease, characterizes by formation of plaque. Coronary endothelial dysfunction is characterized by vasoconstrictive response to the endothelium. Although endothelial dysfunction is considered an early phase of coronary atherosclerosis [1]. It begins with the oxidation of low density lipoproteins (LDL) [2]. Foam cells and smooth muscle cells migrate towards endothelial cell and accumulate to form plaque. Inflammation is not only responsible for development of plaques, but, importantly, plays a crucial role in the destabilization of internal carotid artery plaques, thus converting chronic atherosclerosis into an acute thrombo-embolic disorder. Humoral factors involved in internal carotid artery destabilization include cytokines, cyclooxygenase-2, matrix metalloproteinases, and tissue factor [3]. The T cells then interacts with the macrophages and activates them by releasing tissue factors [4]. These tissue factors rupture the plaque and it activates the platelets and clotting factors [5]. A fibrin net like structure is formed and converts into thrombus formation that leads to the complete blockage and becomes the cause of heart failure. Coronary artery disease is the cause of 52% of incident heart failure in the general population under 75 years [6].

Drugs that are used on present day can cause allergic in our body i.e. swelling of face, lips, tongue, or throat, breathing problem, suddenly severe headache, any abnormal bleeding such as nose bleeds, vaginal bleeding, bloody, stomach pain or discomfort and Nausea. To reduce the observed side effects it would be valuable to propose some new therapeutic targets to treat the inflamed condition. Studies focusing on environmental variables involved in CAD revealed the significant role of *C.pneumonia* in the procured inflammation. *Chlamydia pneumoniae* is a intracellular pathogenic Gram-negative bacterium of the Chlamydiaceae family, that originates an inflammatory process in the pathogenesis of Atherosclerosis [7]. *Chlamydia pneumonia* causing community-acquired pneumonia, bronchitis, sinusi- tis, and upper respiratory tract symptoms. It is known as a leading cause of human respiratory tract infections world-wide. *Pneumoniae* helps in the phenomenon of oxidation of lipoproteins and the induction of cytokines and proteolytic enzyme production, which are essential for Atherosclerosis. *Chlamydia* is responsive for treatment with antibiotics, and the therapeutic effect of the verification of bacterial involvement [8].

2. MATERIAL AND METHODS

2.1 Preparing target molecule

To scrutinize the interaction of ligand, we have taken the predicted RVOM1 protein structure and energy was minimized using Swiss PDB viewer (SPDBV).

2.2 Lead characterization

Three dimensional structure of Stigmasterol was downloaded from PUBCHEM database

(https://pubchem.ncbi.nlm.nih.gov/). Lethal toxicity of these compounds was scrutinized by lazar toxicity prediction server (http://lazar.in-silico.de/predict) on both cell based models and animal models involving Rat and Hamster [9].

3. DOCKING STUDIES

Screening of potential ligand to target protein has been obtained through molecular docking studies [10]. Different poses of interaction of protein and ligand were selected using AutoDock4.2. In order to analyze the effect of ligand association, all the water molecules and the hetero atoms have been removed from the target protein. All the hydrogen atoms were added to the protein as it is required for the electrostatics. Gasteiger charges were assigned to ligands and save them into PDBQT file format [11]. Blind docking has been carried out to predict the highly interacting residues of protein. Best binding pose was determined by minimum binding energy and inhibitory constant through lamarkian algorithm.

4. RESULTS & DISCUSSION

RVOM1 protein is a translocator protein. Gram negative bacteria contain virulent related auto-transporter protein with an N-terminal signal peptide that helps in transportation of protein across the inner membrane [12]. Translocator domain contains β barrel structure that carries the passenger domain to the bacterial surface [13]. There are some chlamydial specific drugs available in the market such as azithromycin, clarithromycin, and levofloxacin. But their dosage and duration is not enough to eradicate chronic *C. pneumoniae* infection [14]. So, there is an urgent need to design some more bacterial specific proteins. Stigmasterol is well established for the inhibition of cholesterol absorption [15]. Thus, this can be used as the possible lead for future drug design against RVOM1 protein.

In the present communication, the lazer toxicity analysis server revealed that this biologically active compound may not be carcinogenic to the animal models such as hamster and mouse. AutoDock was used to carry out the interaction studies between ligand and protein (Fig. 1). Docking studies indicated that stigmasterol may be possible inhibitors of RVOM1 protein with minimum binding energy (-6.45 kcal/mol) as well as inhibitory constant (18.33 μM). Also the QSAR analysis (Table 1) of this compound revealed that the molecular weight and total surface polarity is considerable enough to prevent passage of ligand through the membrane barrier. This non-permeable ligand will interfere with the surface protein (RVOM1) of *C.pneumonia* during the infection. Reduced activity of this protein may lead the pathogenic bacteria to be less virulent and viable. The stigmasterol may be used to create the drug model through the inhibitory effects on bacterial surface protein. Thus, drug targeting this protein will directly interfere with the bacterial auto-transporter function. Thus, it will directly affect the function to carry the proteins or protein domains from the periplasm through the outer membrane to the bacterial cell surface or in the extracellular medium. Present study implicates that stigmasterol may be used as a lead compound for more efficient future drug design through inhibition of RVOM1 protein.

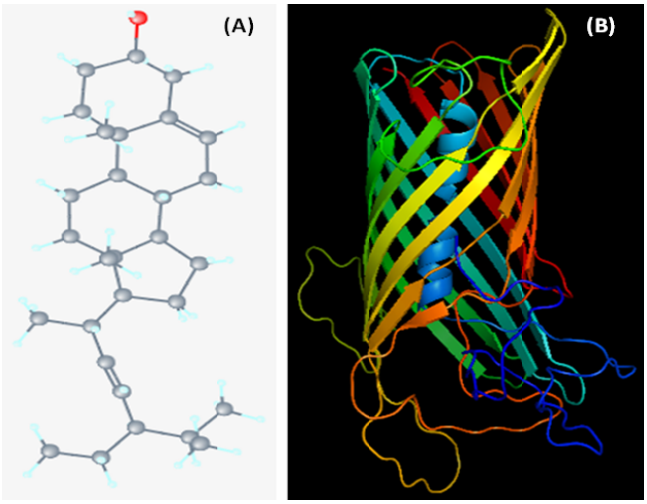

Fig. 1: 3D structure of (A) syigmasterol (B) RVOM1.

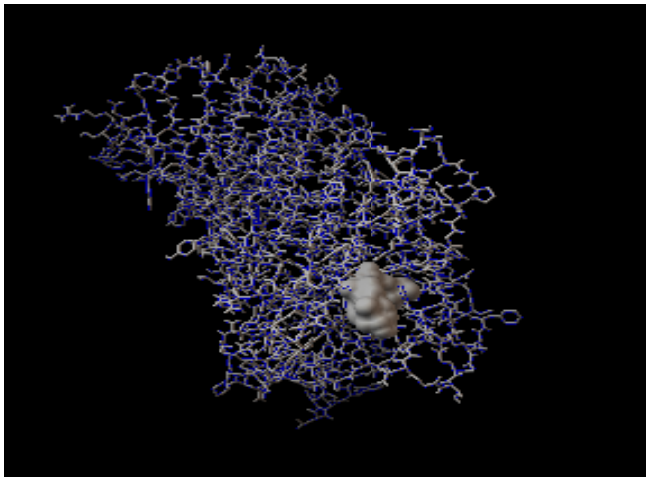

Fig. 1: Docked view of Stigmasterol with RVOM1 protein.

Table 1:- QSAR analysis of Stigmasterol.

S. No.	TPSA*	MW**	LogP	Volume
1.	20.23	412.70	7.87	450.33

***Total Polar Surface Area, ** Molecular Weight**

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