Docking Study of Cysteinyl Leukotriene 1 Receptor: Therapeutic Target for Allergy

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Abstract

Cysteinyl leukotrienes are inflammatory mediators having important role in pathophysiological conditions such as asthma and allergic rhinitis. CysLT1 receptor mediates most of the disease regulatory actions of the CysLTs and it is been implicated in a number of inflammatory conditions including gastrointestinal and cardiovascular diseases. Hence in the present study, molecular docking of CysLT1 was performed with its potent and orally efficacious antagonist CP-199330 and CP-199331. The aim of this study was to compare the interaction of CP-199330 and CP-199331 with known drugs such as Zafirlukast, Pranlukast and Montelukast which had already showed clinical efficacy in the treatment of asthma. The residues such as TYR83, GLN274, LYS311 and SER313 were found to interact with both the antagonist and the known drugs. Also, we noticed the docking scores and interaction of the antagonists were comparable with the known drugs. Hence these antagonists could serve as better drugs for the treatment of allergy.

Keywords: CysLTs, Molecular Docking; CP-199330; CP-199331.

1. Introduction

Cysteinyl–leukotrienes (CysLTs) are formed by the action of 5-lipoxygenase and its activating protein on arachidonic acid and produced by the cells of immune system specially eosinophils, basophils, mast cells and macrophages. The CysLTs are present on outer membrane of inflammatory cells. They are 7-transmembrane G-protein coupled receptors having two subtypes namely CysLT1 and CysLT2 whose seven hydrophobic transmembrane domains are connected by six hydrophilic loops. CysLTs are involved in maturation and migration of dendritic cells and selectively promote the generation of Th2 cytokines and hence enhances allergic responses. CysLTs are important mediators of inflammation and significantly modulate the inflammatory responses seen in asthma and allergic rhinitis. CysLT1 receptor is highly expressed in spleen, peripheral blood leukocytes, interstitial lung macrophages and in airway smooth muscle. Also it has been localized both at gene and protein level in blood vessels and in the interstitial cells. Most of the disease regulatory actions of the CysLTs are mediated through CysLT1. It has been proved that CysLT1 antagonists have a significant role in allergic rhinitis (AR) and asthma and improves pulmonary and lung function, and provide a therapeutic alternative to glucocorticoids in patients with allergic airway disease. The major interest for the development of antagonists of the CysLT1 receptor was because of their beneficial effects in the treatment of asthma, and zafirlukast, montelukast and pranlukast have been clinically introduced for this purpose thus validating the intervention at the CysLT1 receptor as the therapeutic target.

Chambers et al. 1999 have discovered two potent and efficacious CysLT1 antagonist CP-199330 and CP-199331. They proved that these antagonists were equipotent to both zafirlukast and pranlukast in binding to CysLT receptors and showed comparable potency in blocking extracellular calcium influx in human U937 cells. Also, they blocked both antigen and CysLT receptor induced airway obstruction in guinea pigs being equipotent to zafirlukast and pranlukast. The pharmacokinetic profile of these antagonists in rats and monkeys shows low hepatic clearance, moderate terminal
half-life and high oral bioavailability and are devoid of liver toxicity. Hence, in the present study, we aimed to identify the interaction mechanism of two potent Cys-LT1 antagonist CP-199330 and CP-199331 using in silico approach, molecular docking. Though the crystal structure of CysLT1 has not yet been solved, the 3D structure of CysLT1 generated from our previous study has been utilized\[15\]. The Surflex dock module of SYBYL software was used to perform docking. We have created two protomols, a site to perform docking on protein. This was done to analyze whether the antagonist interacts in the same manner as known drugs in different pockets of the protein. The docked mode of the antagonists was obtained and the interaction of the molecules within the protein was clearly understood. We observed that the docking scores and interaction of these antagonists were comparable with the known drugs. Hence these antagonists CP-199330 and CP-199331 could be used for the treatment of allergic disorders.

2. Material and Methods

2.1. Preparation of Protein Structure
The protein structure for docking was prepared using protein preparation tool in biopolymer module of SYBYL. The three dimensional structures of CysLT1 generated from our previous study was used. The models were generated using comparative modeling and threading based approaches. The energy minimization was performed for 100 steps utilizing Tripos force field, Gasteiger Huckel charge and Powell method. The hydrogen molecules and Gasteiger Huckel charge was added to the protein structures during preparation.

2.2. Preparation of Ligand Molecules
The chemical structure of CysLT1 antagonists CP-199330 and CP-199331 were taken from the literature\[14\]. The known drugs zafirlukast, montelukast and pranlukast were downloaded from pubchem database. The antagonists were sketched using sketch molecule

![Chemical structure of antagonist and known drugs for Cysteinyl Leukotriene 1 receptor.](image)

Fig. 1. Chemical structure of antagonist and known drugs for Cysteinyl Leukotriene 1 receptor.
function in SYBYL software\[16\]. The energy minimization of all the molecules was performed using Tripos force field and atomic charges were assigned using Gasteiger Huckel method. The structures of all molecules are shown in Fig. 1.

2.3. Molecular Docking

Molecular docking was performed utilizing Surflex dock module of SYBYL. Two antagonists and three known drugs of CysLT1 were docked into protein CysLT1 receptor. The docking algorithm in surflex dock uses an idealized active site called protomol\[17\]. The protomol is the representation of intended binding site to which the ligand molecules were docked. Two parameters, such as threshold and bloat, determine the extent of a protomol. The protomol was generated using automated mode. Surflex dock uses an empirical scoring function to score the docked ligand conformation which takes into account several terms, including hydrophobic, polar, repulsive, entropic and solvation\[18\].

To evaluate the docking results, the docking scores are expressed in terms of \(-\log_{10}K_d\) units, where \(K_d\) represents a dissociation constant of a ligand.

3. Results and Discussion

3.1. CysLT1 3D Structure Modeling

The three dimensional structure of CysLT1 generated from our previous study\[14\] was utilized to perform docking. The models was predicted using the comparative modeling program, Easy Modeller 4.0 and online threading server I-TASSER and validated using RC plot and ERRAT plot. Based on the validation results the best models (Easy-Modeler: model 8; I-tasser: model 3) were selected because these models has 99.1% and 98.2% of residues in favored and allowed region and 71.6%, and 92.5% overall quality. Therefore these models were selected and used for docking.

3.2. Molecular Docking

Molecular docking was performed for CysLT1 antagonists and the known drugs. Before performing docking the protomol was generated at two different sites of the protein. This was done to verify whether the antagonist interact similar to known drugs at different sites in the protein. When performing docking, 20 different conformations were generated for each molecule and the best

| S.no | Compound name | I-TASSER model | Easy-Modeler model |
|------|----------------|----------------|--------------------|
|      | Docking score | H-bond interaction | Docking score | H-bond interaction |
| 1    | CP-199330     | 6.49 TYR83, GLN274 | 6.18 SER220, LYS311, SER313 |
| 2    | CP-199331     | 7.49 TYR83, GLN175 | 6.73 LYS219, SER313, ARG322 |
| 3    | Montelukast   | 7.23 TYR83       | 7.00 LYS311       |
| 4    | Pranlukast    | 7.58 ARG79, TYR82, TYR83, GLN274 | 7.25 LYS311, SER313 |
| 5    | Zafirlukast   | 7.00 ARG22, TYR83, ARG253 | 5.00 LYS311       |

Fig. 2. Docking mode and interaction of CP-199330 with the CysLT1 receptor.
conformation was chosen based on surflex score and interaction with the residues. The docking score and H-bond forming residues for all the molecules are tabulated in Table 1. We observed that the docking score and H-bond forming residues of CP-199330 and CP-199331 were comparable to the known drugs zafirlukast, montelukast and pranlukast. Also, we noticed, TYR83, LYS311 and SER313 play a major role in interaction of CP-199330 and CP-199331 with the CysLT1 protein. The same interaction was observed in known drugs. This validates that the antagonist could bind efficiently with the protein similar to known drugs. The interaction

**Fig. 3.** Docking mode and interaction of CP-199331 with the CysLT1 receptor.

**Fig. 4.** Docking mode and interaction of Montelukast with the CysLT1 receptor.

**Fig. 5.** Docking mode and interaction of Pranlukast with the CysLT1 receptor.
of antagonist CP-199330 and CP-199331 was depicted in Fig. 2 and 3 respectively. Fig. 4, 5 and 6 shows the docking interaction of known drugs. These results authenticates that CP-199330 and CP-199331 have same binding interaction as known drugs zafirlukast, montelukast and pranlukast which validates these compounds can be used for allergy.

4. Conclusion

In the present study, molecular docking has been performed on two CysLT1 antagonist CP-199330 and CP-199331 and three known drugs zafirlukast, montelukast and pranlukast. The docking results demonstrate that these antagonists docked well within the target protein and interactions were comparable with known drugs. Also, the study clearly indicated the H-bond interaction and binding site of antagonists were similar to the known drugs. Hence, these antagonists could act better in the treatment of allergy.

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