Biomphalaria glabrata RETINOID X RECEPTOR (RXR) PROTEIN ALLOSTERIC SITE IDENTIFICATION BY MOLECULAR MODELING

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Introduction

Schistosomiasis, caused by the parasite *Schistosoma mansoni*, is a leading cause of mortality in tropical and subtropical regions, with the freshwater mollusk *Biomphalaria glabrata* serving as its intermediate host. The disease is directly associated with areas of low human development index (HDI). In this context, retinoic acid nuclear receptors play crucial roles as genetic transcription factors during organogenesis in the embryonic period, influencing the growth of various organisms within the animal kingdom [1]. The retinoid X receptor (RXR) belongs to a superfamily of nuclear receptors that have been extensively studied to identify conserved regions and enhance molecular phylogenetic knowledge. One potential strategy to reduce schistosomiasis incidence involves targeting the mollusk population by controlling the proliferation without harming local biodiversity. This research project aims to utilize molecular modeling to identify allosteric sites of the RXR in *Biomphalaria glabrata* (Uniprot ID: Q8T5C6) which may be applied in the discover of new bioactive allosteric modulators molluscicide, ensuring minimal impact on the RXR of other species [2].

Material and Methods

The RXR proteins considered in this study are from *Biomphalaria glabrata* (Uniprot ID: Q8T5C6), mouse (*Mus musculus,* Uniprot ID: P28700), zebrafish (*Danio rerio,* Uniprot ID: A2T929), and human (*Homo sapiens*, Uniprot ID: P19793) [3]. First, we created a ligand library from the Selleckchem website by downloading files for FDA approved (3,104 compounds), Natural Products (3,057 compounds), Bioactive-1 (9,961 compounds), and Bioactive-2 (5,309 compounds). After optimization of thousands of ligands using the MMFF94 force field (via the Avogadro/OBabel program) and refinement with the PM7 method (MOPAC2016) [4]. The molecular targets of the four species were prepared by comparative modeling (PDB ID: 1XIU) and validation of their three-dimensional structures using the Robetta and SwissModel servers. The Molegro Virtual Docker program [5] was used to adjust the ionization state of amino acid residues and to identify the potential molecular cavities. The primary amino acids sequences of molecular targets and the visualization of residues molecular present in the binding sites were performed using the Pymol program.

Results and Discussion

After geometric refinement of the ligands using semi-empirical quantum calculations (PM7 method), preliminary results showed that the prepared database of thousands of molecules was reduced to a total of 9,931 (47%). Although the individual libraries summed to 21,117, many structures were repeated, and a few were incorrect or missing. For the comparative model, the Ramachandran plot was evaluated, indicating that over 95% of the structures of RXR of different organisms were acceptable. In the orthosteric site, we identified two residues in *B. glabrata* (Gly286 and Thr318) that do not interact with the ligand retinoic acid. However, their equivalents in other species, Ser286 and Ala318, do form

interactions. Conversely, Leu407 interacts with the ligand in *B. glabrata*, but not in the other species. The main differences, however, are observed in the allosteric sites (Figure 1).

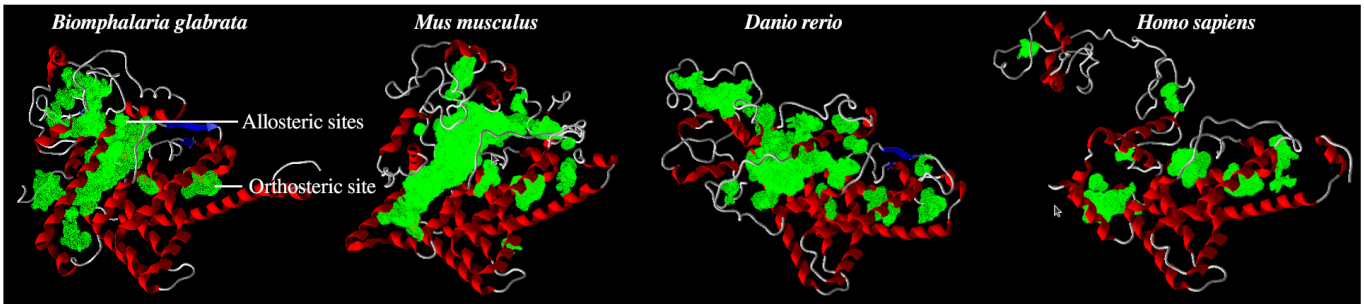

Figure 1. Cavities of RXR are shown in green for each species.

Conclusion

With the areas of low similarity identified in the different protein molecular structures, these potential allosteric sites can be explored in virtual screening analyses to select potential new allosteric modulators of the RXR target.

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