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Studying RhsP2 protein as a new antibacterial toxin targeting RNA

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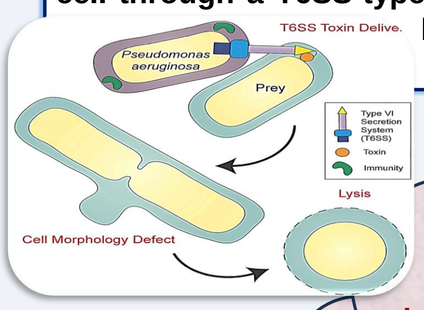
In this research, We tried to shed light on a recent report of Scientists at McMaster University by using a virtual approach to investigate a new potential drug that targets RNAs molecules. disrupts translation of bacterial amino acids to proteins

1. INTRODUCTION & AIM

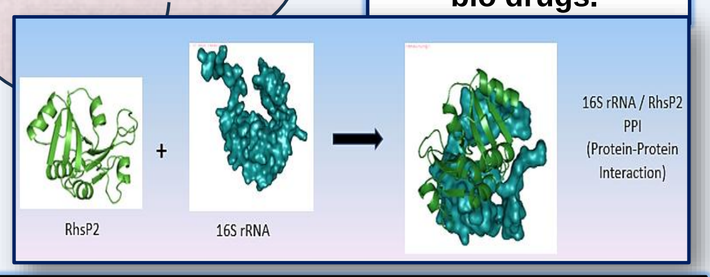
RhsP2 is a toxin secreted by Pseudomonas aeruginosa organisms and is considered one of the group of cytoplasmic enzymes affecting RNA through the mechanism of ADP-ribosyl transferas. This substance enters the other bacterial cell through a T6SS-type VI injection system that delivers the toxin to the other bacterial cells



one can think of this strategy to discover new types of antimicrobials mimic the structure of these bacterial toxins and interact with ncRNAs by different docking software, choose the best hits by using methods and tools of bioinformatics

Aim Of The Work

Identify residues belonging to the active sites of the studied toxin RhsP2 , which target 16S rRNA in order to suggest a novel type of antibacterial bio drugs.



2. Work METHODOLOGY

Using computational docking and modeling, the interactions between two proteins, namely RhsP2 toxin/16S rRNA target, were investigated in detail in order to identify residues belonging to the active sites of the studied toxin RhsP2, which targets 16S rRNA. In our study, we relied on different docking programs such as AutoDock VINA, HADDOCK lite server, and HADDOCK 2.4 server, and compared the results with a reference compound, rifamycin (which works with a mechanism similar to our studied compound), based on criteria related to affinity binding energy and regularity of atoms within the pocket (RMSD).

Steps of work:

1* Using Rifamycin as a ligand to determine the active pockets of 16S rRNA protein, modeled and saved as "pdb" file.

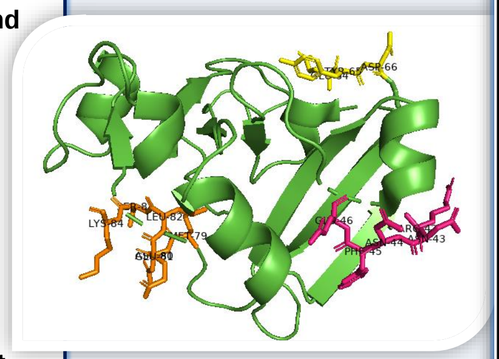
2* Docking the active pocket of 16SrRNA, with RhsP2 (as a ligand and RhsP2 as a protein in stochastic method by AutoDock VINA.

3* Using online docking servers "HADDOCKlite, HADDOCK 2,4" as a protein-protein interaction docking.

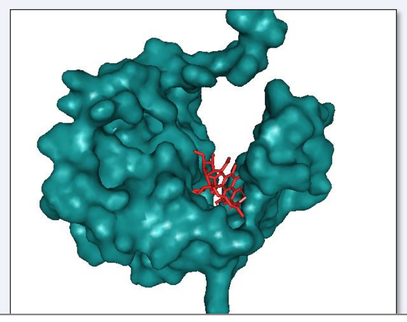
4* After having the docking results, we extracted the active sites of RhsP2 by pyMOL to be the suggested drugs at three sites:

(DRUG1 = GLU-64,TYR-65,ASP-66) , (DRUG2 =ARG-42,ASN-43,ASN-44,PHE-54,GLU-46), (DRUG3 = MET-79,ASN-80,GLU-81,LEU-82,SER-83,LYS-84)

5* Docking each one of extracted pharmacophores by AutoDock VINA to the target receptor, compare their affinity binding energy / RMSD criterion with the reference (rifamycin), for the best conformations structures.

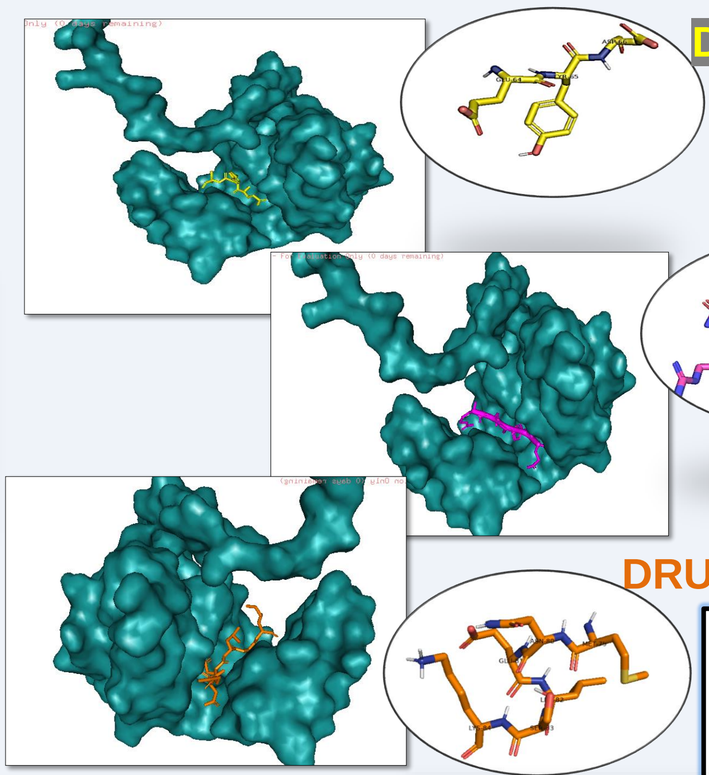


3. RESULTS & DISCUSSION



Active pocket With reference Rifamycin

In order to decide whether the proposed models are good and successful in the biological interaction with the target protein, they must be compared with the standard (Rifamycin) that has certain interference and clear effectiveness when it binds with the active pocket of the target protein 16S rRNA



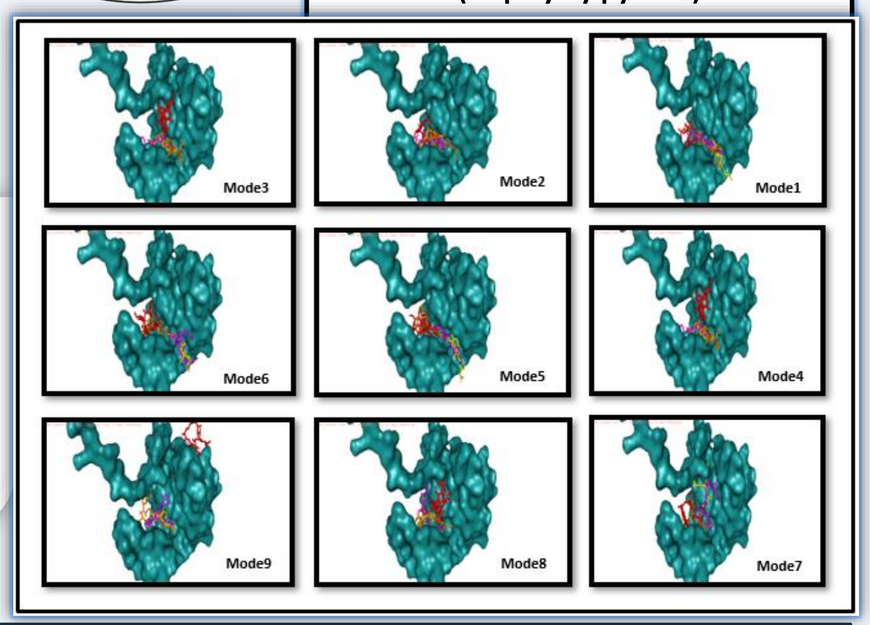
DRUG1

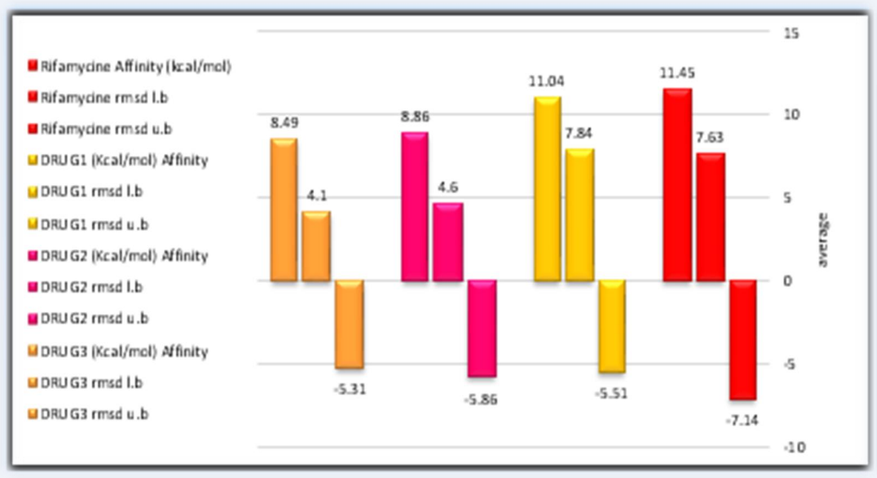
DRUG2

DRUG3

The three potential models of proposed drugs

Nine conformations structures from the three potential drugs (Drug 1,2,3) with the reference in red (display by pyMOL)





4. CONCLUSION

Considering the average affinity binding and RMSD values for the three prodrugs; The results show that the HADDOCK lite server showed the closest model (DRUD2) to rifamycin in interaction with the active pocket of the target protein 16S rRNA, followed by the HADDOCK 2.4 (DRUG3). and finally, the model generated by the Structuring AutoDock VINA method (DRUG1).

Our results were reasonable as a preliminary prediction with one target 16S rRNA, and we can say that the toxic substance RhsP2 has a good interaction with 16S rRNA so we propose it as an inhibitor by binding to the active pocket (such as the action of rifamycin) and inhibiting its function in translating amino acids into proteins.

5. FUTURE WORK / REFERENCES

1. **re-conduct the experimental studies, and docking processes with other human protein targets 18SrRNA, and comparing the new binding scores with the previous 16s rRNA bacterial target in order to determin the effectiveness and toxicity of the studied toxin.**
2. **After a first docking with RNA as a rigid receptor target, it has become increasingly clear that side chain flexibility plays a crucial role in ligand– protein complexes. Therefore, it is necessary to re-conduct the flexible docking.**

1* Chen G, Seukep AJ, Guo M. Recent advances in molecular docking for the research and discovery of potential marine drugs. Mar Drugs. 2020 Oct 30;18(11).

2* First known RNA-targeting toxin launches "total assault" to kill bacteria [Internet]. [cited 2022 Dec 3]. Available from: https://newatlas.com/medical/rna-targeting-toxin-total-assault-antibiotic-bacteria/

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