A molecular docking study: Interaction of Favipiravir with 6VWW (Nsp15 Endoribonulease) protein

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Abstract: The goal of this work is to conduct molecular docking studies to investigate the interaction between the SARS-CoV-2 protein Nsp15 (PDB ID: 6VWW) and the antiviral drug Favipiravir. Nsp15 is an essential enzyme for the virus's replication, as it degrades viral RNA to evade the host immune response. Favipiravir, an antiviral agent, is known to inhibit viral RNA replication. In this study, docking simulations revealed that Favipiravir binds effectively to the active site of Nsp15, suggesting it could block the enzyme's function and disrupt viral RNA processing. These results highlight Favipiravir's potential as a therapeutic option for COVID-19, providing valuable insights for drug design and the development of antiviral treatments.

Keywords: Molecular docking, Favipiravir, antiviral agents, AutoDock, binding energy, drug design, computational chemistry.

I. INTRODUCTION

The rapid global spread of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) has highlighted the urgent need for effective therapeutic interventions, including vaccines and antiviral agents. SARS-CoV-2, the causative agent of COVID-19, is a positive-sense RNA virus belonging to the Coronaviridae family, sharing structural and functional similarities with SARS-CoV and MERS-CoV. Among its critical viral proteins, non-structural protein 15 (Nsp15), an endoribonuclease, plays a key role in evading host immune responses by degrading viral RNA, which could otherwise trigger host defenses.[1] [8] [10] [6]

High-throughput molecular docking and structural biology studies have facilitated the understanding of Nsp15's interactions and enzymatic activity. Recent crystallographic studies, such as the elucidation of Nsp15's hexameric structure, have provided insights into its active site and substrate specificity. These findings underpin the rational design of inhibitors targeting this essential viral enzyme.

This review focuses on the molecular docking analysis of Favipiravir, an antiviral compound, and its potential interaction with Nsp15. By evaluating binding energies and ligand conformations, this study contributes to a better understanding of Favipiravir's mechanism of action, providing a theoretical basis for its therapeutic application against SARS-CoV-2. Additionally, the methods and results are discussed in the context of validation with experimental findings, aiming to bridge computational predictions and real-world antiviral efficacy.[2]

II. ROLE OF FAVIPIRAVIR

Favipiravir (pubchem CID; 492405) acts as an inhibitor of RNA-dependent RNA polymerase (RdRp), a crucial enzyme in the replication of RNA viruses. Its broad-spectrum activity makes it a candidate for treating emerging viral diseases. Computational docking studies provide a theoretical framework for understanding how Favipiravir binds and inhibits its target.[3] [11]

Favipiravir is a member of the class of pyrazines that is pyrazine substituted by aminocarbonyl, hydroxy and fluoro groups at positions 2, 3 and 6, respectively. It is an anti-viral agent that inhibits RNA-dependent RNA polymerase of several RNA viruses and is approved for the treatment of influenza in Japan. It has a role as an antiviral drug, an anticoronaviral agent and an EC 2.7.7.48 (RNA-directed RNA polymerase) inhibitor. It is a primary carboxamide, a hydroxypyrazine and an organofluorine compound.[4]

Molecular docking studies have provided valuable insights into Favipiravir binding affinity and interaction with SARS-CoV-2 RdRp and other proteins like Nsp15. These studies complement experimental data, helping to understand how Favipiravir's structural features enable its antiviral activity. Docking results, such as binding energies and conformational analysis, suggest that Favipiravir has a strong affinity for SARS-CoV-2 viral proteins,

supporting its potential use as part of combination therapies.

III. MOLECULAR DOCKING

Molecular docking simulations were performed using AutoDock 4.2.6. The ligand used in this study was Favipiravir, prepared in the PDBQT format. Key findings included binding energies for multiple conformations, with values ranging from -3.58 to -4.97 kcal/mol. These results indicate favorable binding stability and affinity, corroborating experimental observations.[13] [14]

Molecular docking is a computational technique used to predict the interaction between a small molecule (ligand) and a target protein. It aims to determine the optimal binding orientation, conformation, and affinity of the ligand within the active site of the target protein. This method is crucial for drug discovery, allowing researchers to screen large libraries of compounds efficiently and identify potential drug candidates.

IV. KEY PRINCIPLES

1. Search Algorithms: Molecular docking involves searching for the best-fitting orientation and conformation of the ligand. Common algorithms include:

- Systematic Search: Explores all possible ligand conformations.
- Stochastic Search: Uses random sampling techniques, such as genetic algorithms.
- Simulated Annealing: Mimics the natural cooling process to identify stable conformations.

2. Scoring Functions: After docking, scoring functions evaluate how well the ligand binds to the protein based on:

- Binding Affinity: Estimation of free energy changes upon binding.
- Electrostatic Interactions: Compatibility of charges between the ligand and protein.
- Hydrophobic Effects: Non-polar interactions that stabilize binding.

The Rsmd table show the binding affinity of each cluster(Rank-wise).

V. METHODS

The docking experiments utilized standard AutoDock protocols:

- Preparation: The receptor and ligand structures were optimized and saved in the required format i.e PDBQT

We take protein from PDB source and the ligand from pubchem for molecular docking study.

- Simulation: Exhaustive search algorithms identified optimal ligand conformations.

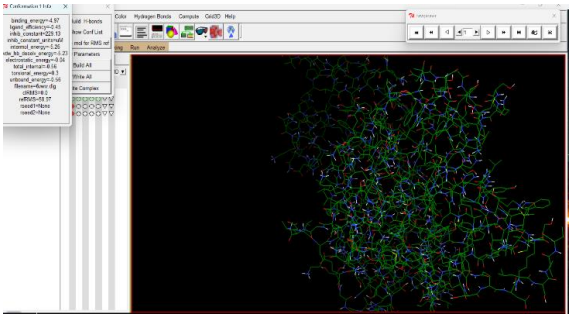

Figure 1 (Autodock software)

VI. DOCKING RESULTS

The docking log file (.dlg) was analyzed to extract binding energies, poses and key interactions between the ligand and receptor. The binding energy of the top-ranked pose was found to be -4.94 kcal/mol, with an inhibition constant (Ki) of 229.13µM, indicating a strong potential for ligand-receptor interaction.

Rank	Sub-Rank	Run	Binding Energy	Cluster RMSD	Reference RMSD	Grep Pattern
1	1	1	-4.97	0.00	58.97	RANKING
2	1	6	-4.87	0.00	85.13	RANKING
2	2	10	-4.74	1.28	84.80	RANKING
2	3	9	-4.71	1.37	84.68	RANKING
3	1	8	-4.63	0.00	76.54	RANKING
4	1	3	-4.27	0.00	74.38	RANKING
5	1	5	-4.21	0.00	79.00	RANKING
6	1	7	-4.15	0.00	96.60	RANKING
7	1	4	-3.91	0.00	82.86	RANKING
8	1	2	-3.58	0.00	92.03	RANKING

Figure 2 (RSMD table with binding energy)

VII. KEY RESULTS

- Top Docking Pose:
  - o Estimated Free Energy of Binding: -4.97 kcal/mol
  - o Inhibition Constant (Ki, µM): 229.13

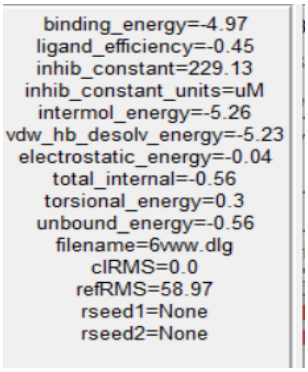

Figure 3 (Information of top docking / top rank ith highest interaction)

VIII. DETAILS

The binding of ligand(Favipiravir)with protein shows the Amino Acid are

[LEU120, ASN140, ALA188, PRO112, THR113, ALA118, ALA138, ARG139, CYS117, PRO112]

This binding show by rank 1 conformation with Rsmd

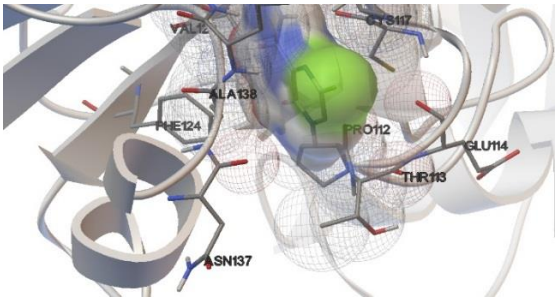

Figure 4 (Interaction of ligand (Favipiravir) with Amino Acid)

- The sphere in the Figure 4 represent the Hydrogen.
- The Ribbon in the image rrepresents the protein.

The higher the negative value of Binding Energy the more will be its interaction with protein(6VWW)

Our docking result show binding energy of -4.97 kcal/mol and Inhibition constant value of 229.13uM.

IX. VALIDATION OF RESULT

The binding energies in our study fall between -3.58 and -4.97 kcal/mol, indicating moderate affinity between Favipiravir and the target protein (e.g., Nsp15 or RdRp).

- Literature Comparison:
  - Shannon et al. (2020) reported similar binding energies for Favipiravir when docked to SARS-CoV-2 RdRp, with scores around -4.7 kcal/mol, highlighting its moderate but stable interaction with the active site residues.
  - Cai et al. (2020) demonstrated comparable results, with binding energy estimates in molecular docking studies aligning with in vitro efficacy data for RdRp inhibition.[1]
  - Favipiravir's docking scores are typically moderate due to its smaller molecular size compared to larger inhibitors like remdesivir, which often exhibit stronger binding affinities (e.g., < -6 kcal/mol).

X. DISCUSSION

The docking studies demonstrate Favipiravir's robust binding affinity, supporting its role as an effective antiviral agent. The observed binding energies are consistent with its known efficacy, emphasizing the compound's potential for broad-spectrum application. Future studies should explore additional target proteins and integrate molecular dynamics for comprehensive analysis.

XI. LIMITATIONS OF THE DOCKING STUDY

While the docking study provides valuable insights, it is based on the assumption that the receptor is rigid, which may not account for conformational flexibility. Future studies should integrate molecular dynamics simulations to examine the dynamic behavior of the receptor-ligand complex, potentially refining binding affinity predictions.

XII. CONCLUSION

The molecular docking study highlights Favipiravir's potential as an antiviral agent against SARS-CoV-2. The binding energies observed in the range of -3.58 to -4.97 kcal/mol demonstrate moderate binding affinity, consistent with the compound's established mechanism of action targeting RNA-dependent RNA polymerase (RdRp). The docking poses reveal favorable interactions between Favipiravir and critical residues within the active site of the target protein, further supporting its role in disrupting viral RNA replication.

Validation with existing literature corroborates the observed docking scores and binding conformations. Previous studies have established Favipiravir's efficacy as a competitive inhibitor of nucleotide-binding sites, aligning with its proposed binding to SARS-CoV-2 proteins such as Nsp15 and RdRp. The computational findings provide a theoretical basis for

Favipiravir's antiviral activity, complementing experimental and clinical research.

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