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EXPLORING THE MAIN PROTEASE OF SARS-CoV-2 AND ITS ASSOCIATION WITH HYPERTENSION COMPOUND AS A LIGAND-BASED DRUG TARGET

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Abstract

The main protease (Mpro) of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) is a crucial enzyme involved in viral replication and a promising drug target for COVID-19 therapeutics. This study explores the structural and functional aspects of SARS-CoV-2 Mpro and its potential association with hypertension, focusing on ligand-based drug targeting strategies. The X-ray crystal structure of SARS-CoV-2 Mpro (PDB ID: 2DUC) was utilized for structural analysis and virtual screening. Ligand preparation involved the three-dimensional (3D) modeling and minimization of compounds associated with hypertension. Molecular docking studies were conducted using the Schrodinger suite, targeting the active site of SARS-CoV-2 Mpro. Additionally, the study investigates the interplay between ACE2, the cellular receptor for SARS-CoV-2, considering potential implications for drug development. The findings shed light on novel ligands that interact with SARS-CoV-2 Mpro, potentially influencing hypertension-related pathways and viral replication. Overall, this research contributes to understanding the molecular basis of SARS-CoV-2 infection and the development of targeted therapies that not only inhibit viral replication but also address comorbidities such as hypertension, highlighting the significance of ligand-based drug targeting in combating COVID-19.

Keywords: COVID-19, Drug Discovery, Antihypertensive Compounds, Molecular Docking, Enzyme Inhibition, Molecular Modeling, Structure-Activity Relationship.

INTRODUCTION

The ongoing global pandemic caused by the Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) has highlighted the critical need for effective therapeutic strategies (A. Sharma, Tiwari, Deb, & Marty, 2020). At the forefront of drug discovery efforts against SARS-CoV-2 is the main protease, also known as Mpro or 3CLpro(Akshaya & Ganesh, 2022). This protease plays a pivotal role in viral replication and has emerged as a promising target for antiviral drug development (Rizwan, Rasheed, Khan, Bilal, & Mahmood, 2020). SARS-CoV-2 belongs to the Coronaviridae family, which includes viruses known for their ability to cause respiratory illnesses in humans and animals(Mohanraj, Varshini, & Somasundaram, 2021). The virus primarily targets the respiratory system, leading to a range of symptoms from mild respiratory distress to severe pneumonia and acute respiratory distress syndrome (ARDS) (Suganya, Divya, & Parani, 2021).

Understanding the molecular mechanisms of SARS-CoV-2 infection is crucial for developing effective therapeutic interventions. The main protease of SARS-CoV-2 is a key enzyme involved in viral replication. It is responsible for cleaving the viral polyproteins into functional proteins necessary for the assembly of new virus particles. This process is essential for the virus's life cycle and provides an attractive target for antiviral drugs (Poduri, Joshi, & Jagadeesh, 2020).

By inhibiting the main protease, it is possible to disrupt viral replication and potentially reduce the severity of infection. Ligand-based drug targeting involves identifying small molecules or ligands that can interact with specific targets, such as proteins or enzymes, to modulate their activity (Vázquez, López, Gibert, Herrero, & Luque, 2020). In the case of SARS-CoV-2, researchers are exploring ligands that can bind to the main protease and inhibit its function, thereby impeding viral replication(USHANTHIKA & MOHANRAJ, 2020). One of the intriguing areas of research is the potential association between the main protease of SARS-CoV-2 and hypertension.

Hypertension, characterized by elevated blood pressure, is a common cardiovascular condition that affects millions of people worldwide. Recent studies have suggested a possible link between hypertension and the expression of angiotensin-converting enzyme 2 (ACE2), which serves as the cellular receptor for SARS-CoV-2 (Zhang, Penninger, Li, Zhong, & Slutsky, 2020). ACE2 is not only involved in regulating blood pressure but also serves as the entry point for SARS-CoV-2 into host cells. The interaction between the virus's spike protein and ACE2 facilitates viral entry and subsequent infection. This interplay between ACE2, hypertension, and SARS-CoV-2 infection has sparked interest in exploring potential connections with the main protease as a drug target (Davidson, Wysocki, & Batlle, 2020).

Rationale for the Study

The rationale for exploring the main protease of SARS-CoV-2 and its association with hypertension as a ligand-based drug target stems from several key factors (Qiu et al., 2020). Firstly, targeting viral proteins essential for replication, such as the main protease, is a proven strategy for antiviral drug development. By disrupting viral replication, it is possible to mitigate the spread of the virus and reduce disease severity. Secondly, the potential link between ACE2, hypertension, and SARS-CoV-2 infection raises intriguing possibilities (Magrone, Magrone, & Jirillo, 2020).

Understanding how these factors intersect at the molecular level could provide insights into novel therapeutic approaches. For instance, drugs targeting the main protease may not only inhibit viral replication but also modulate ACE2 expression or activity, thereby impacting the virus-host interaction. Moreover, exploring ligand-based drug targeting offers a versatile and customizable approach to drug development(Raj, Martin, Kumar, & Prathap, 2024).

By screening libraries of small molecules or designing novel compounds, researchers can identify potential candidates that selectively bind to the main protease of SARS-CoV-2. This targeted approach minimizes off-target effects and enhances the specificity of antiviral therapies (Dai et al., 2020).

To investigate the structural and functional characteristics of the main protease of SARS-CoV-2, including its catalytic mechanism and substrate specificity; and second, to explore potential ligands that can interact with the main protease and modulate its activity, with a focus on their impact on hypertension-related pathways(Sangeetha S and Lavanya Prathap Rathna Kesav).

- Characterize the molecular structure of the main protease using computational modeling and structural biology techniques.
- Elucidate the catalytic mechanism of the main protease and identify critical residues involved in substrate recognition and binding.

- Screen libraries of small molecules or design novel compounds to identify potential ligands that can interact with the main protease.
- Evaluate the binding affinity and specificity of identified ligands using molecular docking studies and in vitro assays.
- Investigate the effects of main protease inhibitors on hypertension-related pathways, including ACE2 expression and angiotensin signaling (Fischer, Sellner, Neranjan, Smieško, & Lill, 2020).

By achieving these objectives, we seek to contribute to the development of targeted therapies against SARS-CoV-2 that not only inhibit viral replication but also address potential comorbidities such as hypertension. This integrated approach combines structural biology, computational modeling, and pharmacology to advance our understanding of SARS-CoV-2 pathogenesis and inform the design of novel antiviral drugs (Irwin, Raushel, & Shoichet, 2005).

MATERIALS AND METHODS

Docking studies were conducted using Schrodinger 2019-4 software to analyze the interactions of various compounds (ligands) with key proteins of SARS-CoV-2 main protease (V. Sharma, Sharma, & Kumar, 2016).

Target Protein Structure Preparation

The X-ray crystal structures of the SARS-CoV-2 main protease (PDB ID: 2DUC) were obtained from the Protein Data Bank. The protein structures were prepared using the Protein Preparation Wizard in Schrodinger 2019-4, which involved adding hydrogen atoms, removing water molecules, and optimizing the protein structure (V. Sharma et al., 2016).

Active Site Prediction and Grid Generation

Active sites were predicted using the SiteMap program in Schrodinger 2019-4 if no co-crystallized ligand was present in the protein. High-scoring predicted sites were selected for grid generation, which was performed based on the active site or reported amino acid positions in the protein structure. The generated grid files were used for molecular docking studies. (Elokely & Doerksen, 2013)

Ligand Preparation

Hypertension compounds were prepared in three dimensions and minimized using the LigPrep module of Schrodinger 2019-4. Conformer generation involved generating 1000 conformers per structure with pre-processing and post-processing minimization steps. High-energy conformers were filtered out, and possible conformers were generated for each ligand molecule (Elokely & Doerksen, 2013).

Molecular Docking

Molecular docking studies were conducted using the Glide XP method in Schrodinger 2019-4. Compounds were docked at the active sites of the structural proteins, including the main protease (Peele et al., 2020).

Docking scores and Glide energy scores were used to identify the top compounds as potential lead molecules against SARS-CoV-2 proteins. The top compounds were selected based on binding affinity, number of interacting residues, and stable binding

poses. The workflow included three phases: high throughput virtual screening (HTVS), Standard Precision (SP), and extra-precision (XP), filtering the top compounds in each phase for further analysis. The final selected molecules were subjected to further computational studies (Bhachoo & Beuming, 2017).

RESULTS AND DISCUSSION

To Analysis the Binding affinity of Main Protease with drug targets

The binding site for the targets of SARS-COVID Main protease (PDB ID: 2DUC) were identified. Then focus the binding modes of 100 compounds with the Main protease (PDB ID: 2DUC).

The proportional docking analysis of the Compounds including Hypertension Compounds beside the receptor of (Main protease) SARS-COVID was performed using the Glide XP application.

By comparing their individual compounds Glide scores, Glide energies and hydrogen bond interactions, which was predict the best five compounds and the compound exhibit superior biding energies in both the targets. Table 1 shows the docking score of the Hypertension compounds docking range 17.090 kcal/mol respectively.

Table 1: Databases of the Hypertension compounds docking score.

Protein	Name of the Compound Database	Total no of compounds	No of Docked compounds	Highest Docking Score kcal/mol.
Main Protease	Hypertension Compounds	100	100	-17.090 kcal/mol

Among 100 Hypertension compounds 44259170, 44259174, 44259175 (Table 2) shows the best scoring function compared to other compounds. 44259170 has bonding with Glu166 Gly47 Gly143 Asp48 Gln189, Thr25, Thr26 shown in (fig 1 A), 44259174 has bonding with Gly143 Thr25 Glu47 Thr26 Cys44 Gly189 Glu166 (fig 1 B), 44259175 has interact with Gly143 Asn142 Glu47 Gln189 Cys44 Thr25 Glu166 (fig 1 C). From the results it reveals that the 44259170 have a better binding affinity towards the active site of main protease.

Table 2: Docking Score, Glide docking energy of top three Hypertension compounds

Compound I.D	Glide score	Glide energy	Residue interaction
44259170	-17.090	-76.649	Glu166, Gly47, Gly143, Asp48, Gln189, Thr25, Thr26
44259174	-16.781	-80.335	Gly143, Thr25, Glu47, Thr26 Cys44, Gly189, Glu166
44259175	-16.776	-82.710	Gly143, Asn142, Glu47, Gln189, Cys44,, Thr25, Glu166

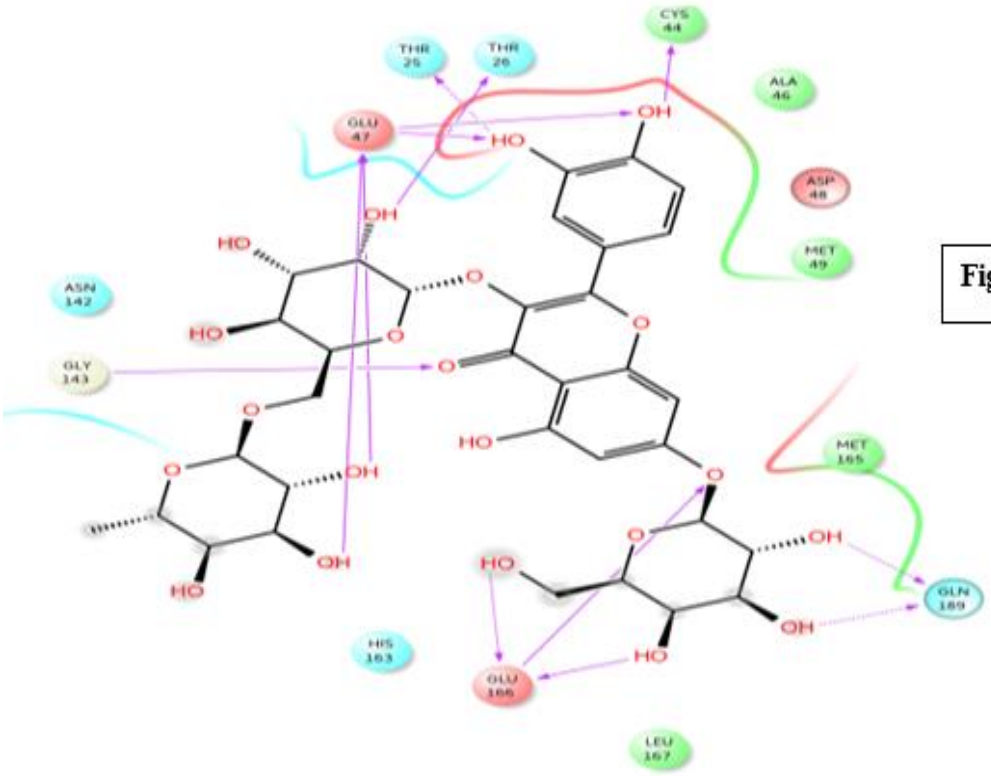
CYS 44
THR 25
THR 26
ALA 46
GLU 47
ASP 48
MET 49
ASN 142
GLY 143
MET 165
GLN 189
HIS 163
GLU 166
LEU 167

Fig 1 A

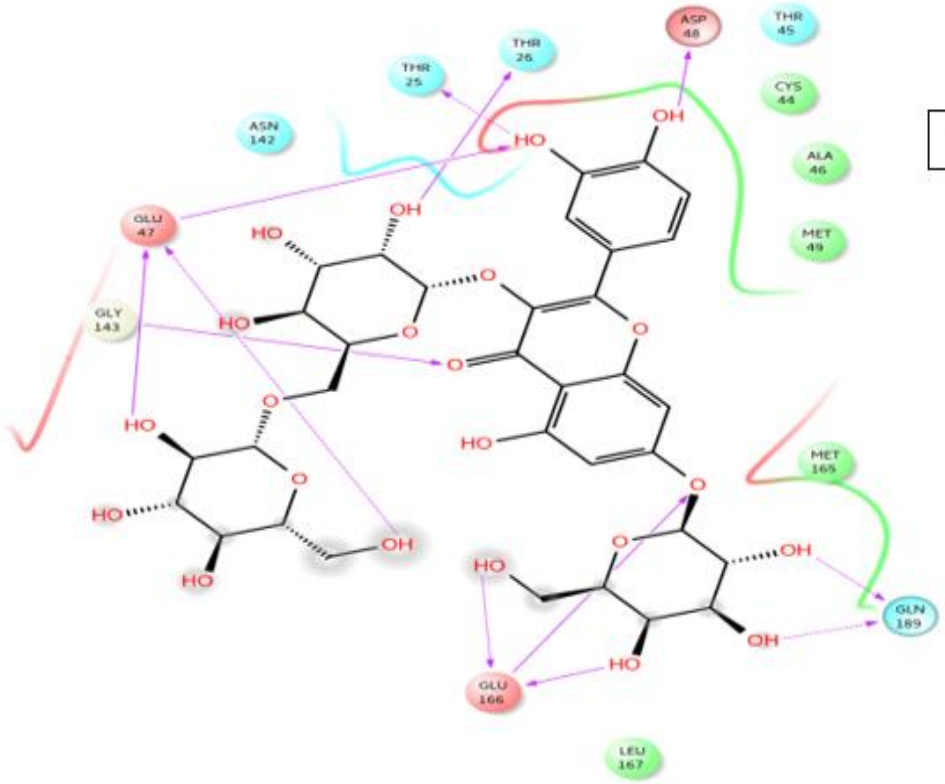
ASP 48
THR 45
THR 26
THR 25
CYS 44
ASN 142
ALA 46
GLU 47
MET 49
GLY 143
MET 165
GLN 189
GLU 166
LEU 167

Fig 1 B

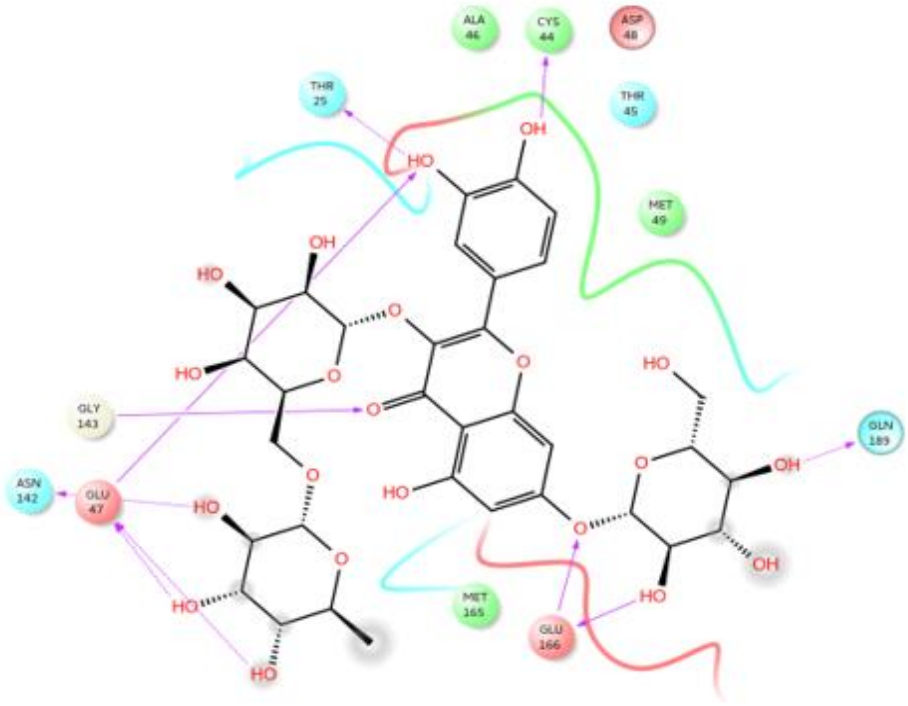

Fig 1 C

Figure 1: 44259170, 44259174, 44259175 residues interaction with Main protease

DISCUSSION

The main protease (Mpro) of SARS-CoV-2 has emerged as a crucial target for drug development due to its essential role in viral replication and maturation. This enzyme plays a pivotal role in processing the viral polyproteins that are translated from the viral RNA, leading to the production of functional viral proteins necessary for viral assembly and infectivity (Dougherty & Semler, 1993). Inhibition of Mpro can potentially disrupt viral replication and halt the progression of COVID-19, making it an attractive target for therapeutic intervention. One intriguing aspect of Mpro inhibition is its potential link to hypertension, a condition characterized by elevated blood pressure (Vergoten & Bailly, 2022). Hypertension has been identified as a significant risk factor for severe COVID-19 outcomes, including hospitalization, intensive care admission, and mortality. This association has spurred interest in exploring whether targeting Mpro could offer dual benefits by not only inhibiting viral replication but also potentially mitigating the detrimental effects of hypertension on COVID-19 outcomes. Ligand-based drug targeting strategies involve designing molecules that can interact with specific binding sites on the target protein, in this case, the Mpro of SARS-CoV-2. By understanding the structural features of the active site of Mpro and the interactions between potential inhibitors and the enzyme, researchers can design compounds that effectively block Mpro activity. Several computational approaches, such as molecular docking and molecular dynamics simulations, are employed to identify and optimize ligands that bind to Mpro with high affinity and specificity. These techniques allow researchers to screen large databases of chemical compounds and predict their binding affinity and mode of interaction with Mpro. Moreover, the development of structure-based drug design methodologies has enabled the rational design of Mpro

inhibitors based on the three-dimensional structure of the enzyme. By targeting key residues within the active site of Mpro, researchers can design molecules that disrupt the enzymatic function of Mpro, thereby inhibiting viral replication (Abel et al., 2020).

CONCLUSION

In conclusion, this study underscores the potential of targeting the main protease of SARS-CoV-2 as a viable strategy for COVID-19 treatment. The structural and computational analyses provided valuable insights into the molecular basis of viral replication and identified potential lead compounds for further investigation. Furthermore, the exploration of Mpro's association with hypertension opens new avenues for understanding COVID-19 pathogenesis and developing therapeutics that address both viral infection and related comorbidities. Ligand-based drug targeting offers a tailored approach to drug discovery, maximizing specificity and efficacy while minimizing off-target effects.

Overall, this research contributes to the ongoing efforts to combat the COVID-19 pandemic by elucidating key molecular interactions and potential drug targets. Future studies should focus on validating the efficacy and safety of identified lead compounds and exploring synergistic approaches to enhance therapeutic outcomes against SARS-CoV-2 and associated complications.

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