Beclabuvir can inhibit the RNA-dependent RNA polymerase of newly emerged novel coronavirus (SARS-CoV-2)

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Abstract

Recent emergence of novel coronavirus (SARS-CoV-2) all over the world has resulted more than 33,106 global deaths. To date well-established therapeutics modules for infected patients are unknown. In this present initiative, molecular interactions between FDA-approved antiviral drugs against the Hepatitis-C virus (HCV) have been investigated theoretically against the RNA-dependent RNA polymerase (RdRp) of SARS-CoV-2. HCV and SARS-CoV-2 are both +ssRNA viruses. At 25° C beclabuvir, a non-nucleoside inhibitor of the $RdRp_{HCV}$ can efficiently bind to RdRp $_{SARS-CoV-2}$ ($\Delta G_{AutoDock}$ = -9.95 kcal mol^{-1}) with an inhibition constant of 51.03 nM. Both the ΔG_{London} and $\Delta G_{GBVI/WSA}$ values were - 9.06 and - 6.67 kcal mol^{-1}, respectively for binding of beclabuvir to $RdRp_{SARS-CoV-2}$. In addition, beclabuvir has also shown better binding free energy with $RdRp_{SARS-CoV-2}$ (ΔG_{vina} = -8.0 kcal mol^{-1}) than that observed with the Thumb 1 domain of $RdRp_{HCV}$ (ΔG_{vina} = -7.1 kcal mol^{-1}). InterProScan has suggested the RNA-directed 5'-3' polymerase activity exists within 549th to 776th amino acid residues of $RdRp_{SARS-CoV-}$, where the major amino acid residues interacting being I591, Y621, C624, D625, A690, N693, L760, D762, D763 and E813-N817. Molecular interaction suggests occupancy of beclabuvir inside the active site environment of the $RdRp_{SARS-CoV-2}$, the enzyme essential for viral RNA

synthesis. In conclusion, results suggest beclabuvir may serve as an anti-SARS-CoV-2 drug.

Keywords

Novel coronavirus (SARS-CoV-2), RdRp, HCV, beclabuvir, *in silico*, molecular docking

Introduction

The newly emerged novel coronavirus (SARS-CoV-2) was first reported from patients with connection history at the Wuhan Seafood market in Hubei province of China in December 2019. Clinical symptoms of the infection caused by this newly arrived pneumonia virus were similar to the previously emerged severe acute respiratory syndrome coronavirus (SARS-CoV) and Middle East respiratory syndrome coronavirus (MERS-CoV) infections [1]. As of 30th March, 2020, confirmed 6,93,224 positive cases have been registered with 33,106 global deaths out of which 3,310 deaths occurred in China and 10,781 deaths in Italy [2]. The super-spreader feature of SARS-CoV-2 is continuously increasing the confirmed positive cases and so far the estimated global death rate is 4.77 % [2]. Social distancing and travel restrictions have been implemented globally as non-pharmaceutical measures to control the fire of the outbreak [3]. In the beginning, governments of different nations evacuated their citizens from China and initiated quarantine protocols to save them. In certain serious areas of China and Italy citizens have been locked-down in their homes as they are at the fourth stage of the Pandemic [4]. Due to the severity of virus outbreak, the World Health Organisation (WHO) has declared this new novel coronavirus outbreak as a Pandemic [5]. Flowing China, Italy has now become the new epicenter of SARS-CoV-

2 infection where 97,689 positive cases and 10,781 (11.03 %) deaths of SARS-CoV-2 infection have been recorded with in a few weeks [6].

In the South-East Asian region, India ranks as the third most-affected country after Indonesia and Thailand respectively [2]. India, has reported 1071 confirmed positive cases and 29 deaths from SARS-CoV-2 infection as of 30th March 2020. Maharastra, the state of India, has become the most affected state, which indicates its vulnerbility to infectious diseases. Patients with an international travel history and symptoms of viral-flu were suspected and tested with expensive SARS-CoV-2 test costing nearly $67. The respiratory illness caused by this new SARS-CoV-2 collectively called, coronavirus disease 2019 (COVID-19) [2]. In the very beginning of 2020, and the new decade, COVID-19 has impacted on the global economy and resulted in a near shutdown of the global equity market, for example on 19th February 2020 Tesla, Inc. ended the day down by 17 %, which is the second-worst drop ever. The global airline industry has also been badly knocked-down by the COVID-19 global Pandemic.

Coronavirus is a member of a large virus family, the coronaviridae and subfamily orthocoronavirinae which usually causes respiratory illness to humans and animals [7]. Soon after the first report of the outbreak, a research group lead by Dr. Zhang at Fudan University, China has sequenced the complete genome which is nearly 30,000 nucleotides long and encodes 10 proteins (MN908947.3), [8]. Like SARS-CoV, the newly emerged novel coronavirus (SARS-CoV-2) is about 60 -140 nm in diameter and has trimeric spike glycoprotein (S), membrane capsid protein (M), small membrane protein, viral nucleoprotein (N) and an RNA dependent RNA polymerase (RdRp), which are essential for virus replication [9]. Usually, entry of human coronavirus is mediated by interactions of spike glycoprotein with angiotensin

converting enzyme 2 (ACE2), aminopeptidase N (AmoPep-N) and dipeptidyl peptidase 4 (DPP-4), [10, 11]. The new SARS-CoV-2 is phylogenetically closer to known coronavirus strains of bat-SL-CoVZC45 and bat-SL-CoVZXC21 than SARS-CoV [12].

Research has suggested nonstructural proteins (NSPs) and structural proteins (SPs) of SARS-CoV-2 have linked a function. For example, NSP can block the host innate immune response and the viral envelope promotes viral assembly and release [13]. However, optimal therapeutic drug for treating the infected patients are still unknown. In the entire genome of SARS-CoV-2, 93 mutations were detected out of which 29 mis-sense mutations were found in the ORF1ab polyprotein that encodes the RdRp [13]. SARS-CoV-2 and HCV, are both +ssRNA viruses and have 22.22 % sequence similarities between their RdRps. In this study, an attempt has been made to identify the most efficient inhibitor of $RdRp_{SARS-CoV-2}$ with the help of computational approaches and by screening the inhibitory properties of commonly used $RdRp_{HCV}$ (NS5B) inhibitors.

Methods

Data sampling and analysis

The nucleotide sequences of SARS-CoV-2 were originally retrieved from NCBI-Virus, Accession Number: MN908947.3. Multiple sequence alignment was performed using MEGA-v2016, [14]. The amino acid sequence of the viral RdRp was scanned for identification of known functional domains/motifs using InterPro Scanner [15]. Homology modeling of the RdRp was carried out using SwissModel and quality was checked using PROCHECK [16]. Rigid-body molecular docking between several

established antiviral drugs and $RdRp_{SARS-CoV-2}$ was carried out using AutoDock (v4.2), [14]. Additionally, implicit solvation was performed using Molecular Operating Environment (v9.10) [14]. Molecular docking studies were visualized using PyMol [14]. Molecular interactions of different antiviral drugs *i.e.*, beclabuvir, tegobuvir, dasabuvir, deleobuvir, setrobuvir, radalbuvir, lomibuvir, remdesivir, uprifosbuvir and favipiravir were investigated with the $RdRp_{SARS-CoV-2}$.

Results

The amino acid sequences of the viral RdRp were compared by multiple sequence alignment in order to understand the evolutionary difference among with previously reported coronavirus strains. Dissimilarities were prominent among coronavirus strains isolated from a different source of host organisms (Figure 2). The RdRp (PDB:6NUR) of previously emerged SARS-CoV was used as a template in obtaining the homology model of the RdRp [17]. The homology model of RdRp showed 96.35 % sequence similarities with 6NUR. The GMQE and QMEAN were 0.83, and -0.71, respectively, where 98.1 % of the amino acid residues were within Ramachandran favored zone which combinedly suggest the high quality of the homology model. Molecular docking studies are summarised in Table 1. Data presented in Table 1 suggest thermodynamically favoured binding properties of beclabuvir and other antiviral drugs with the RdRp of SARS-CoV-2. Among all the aniviral drugs studied beclabuvir showed the best $\Delta G_{AutoDock}$ of -9.95 kcal mol^{-1} followed by tegobuvir (-9.68 kcal mol^{-1}) and dasabuvir (-9.38 kcal mol^{-1}). However, best ΔG_{London} was found for lomibuvir (-10.96 kcal mol^{-1}) followed by setrobuvir (-10.52 kcal mol^{-1}) and dasabuvir (-10.38 kcal mol^{-1}). Setrobuvir also displayed best $\Delta G_{GBVI\,/}$

$_{WSA}$ (-7.48 kcal mol^{-1}) followed by remdesivir (-7.36 kcal mol^{-1}), radalbuvir (-7.36 kcal mol^{-1}) and dasabuvir (-6.68 kcal mol^{-1}). Allthough, lowest inhibition constant is offred by remdesivir (18.12 nM) but, best $\Delta G_{AutoDock}$ (-9.95 kcal mol^{-1}) and inhibition constant (51.03 nM) is offered by beclabuvir with ΔG_{London} and $\Delta G_{GBVI / WSA}$ of -9.06 kcal mol^{-1}, -6.67 kcal mol^{-1} respectively (Table 1). Currently, WHO is conducting a clinical trial program, called global "Solidatory Trial", in which only remdesvir is included.

Discussion

Upon entry inside the host cell, viral RdRp enables multiplication of the +ssRNA virus [18, 19] and eventually the consequences of the viral infection in the host [17]. Thus, RdRp has always been a favorite drug target in antiviral therapeutic research [20]. Nowadays, different antiviral strategies such as, capping the virus particle which enables tethering it into the host epithelial cell membrane [21], designing a DNA vaccine against the major glycoproteins displayed outside the viral capsid protein [22], targeted delivery of miRNA [23], combination antiviral drug therapy [24], etc are commonly employed for therapeutic purposes. Results of the present study suggest that ATP binding domain and RNA-directed 5'-3' polymerase activity of the RdRp exist within 549th to 776th amino acid residues (GO:0003968) and this stretch of amino acids is crucial for RNA synthesis (Figure 3). Beclabuvir is a non-nucleoside polymerase inhibitor that potentially inhibit nonstructural protein 5B (NS5B) of HCV [25]. Previous studies, including phase 3 clinical trials have suggested beclabuvir have a high response rate at post-treatment week [26, 27]. Moreover, the pharmacokinetic, efficiency and tolerability were also reported very favorable [28]. Upon intracellular uptake beclabuvir allosterically binds to the non-catalytic Thumb 1 site (Figure 4) of

viral RdRp which slowdown the RNA synthesis process [25]. However, in the case of RdRp$_{SARS-CoV-2}$ beclabuvir occupies the active site environment (ASE), (Figure 5) is essential for the RNA-dependent polymerase activity [18, 19]). The major interacting residues were I591, Y621, C624, D625, A690, N693, L760, D762, D763 and E813-N817 (Figure 1). Moreover, beclabuvir also showed better binding free energy (ΔG_{vina} = -8.0 kcal mol^{-1}) for RdRp$_{HCV}$ (ΔG_{vina} = -7.1 kcal mol^{-1}). Properties of molecular interaction of beclabuvir and other FDA approved antiviral drugs against HCV infection are summarized in Table 1 which could also be effective to use against SARS-CoV-2 infection. This preliminary study may be contributory towards the anti-epidemic efforts against the newly emerged SARS-CoV-2 [9, 29-31] and may help to shorlist antiviral drugs against SARS-CoV-2 for clinical trials.

Conclusion

Throughout history, the human race had been challenged by multiple of global-pandemics. However, with the advancement of medical science and at the age of Artificial Intelligence, global health disaster of this scale, probablly would not be difficult to manage. However, the things that matter most are the time and the supply of resources. As reported by WHO, the original outbreak began in December-2019 and within three months, global death toll due to SARS-CoV-2 has reached the tragic milestone of 26,000. In conclusion, this rapid study suggests beclabuvir, and other commonly used antiviral drugs for HCV could be utilized as anti-SARS-CoV-2 drugs to control the devastation of the outbreak. However, more studies are required before the practical use of these antiviral drugs.

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Declaration of interests

The author declares no competing financial interests.

Figure Legends

Figure 1. Molecular interaction between beclabuvir and RNA dependent RNA polymerase of newly emerged novel coronavirus (RdRp$_{SARS-CoV-2}$). **A.** Major interacting amino acid residues which occupy the catalytic site of RdRp$_{SARS-CoV-2}$. **B.** Binding pose of beclabuvir with RdRP$_{SARS-CoV-2}$.

Figure 2. Phylogenetic position of the RdRp$_{SARS-CoV-2}$. Tree was constructed using multiple sequence alignment followed by neighbor-joining method with 1000 bootstraps replicates. The alpha-numeric values at right indicate Protein Data Bank accession ID. The numeric values at the top of the tree branch indicate bootstraps replicate values.

Figure 3. Protein domain/motif analysis of the RdRp$_{SARS-CoV-2}$. The yellow highlighted region indicates the catalytic site (RNA-directed RNA-polymerase activity) of the RdRp$_{SARS-CoV-2}$.

Figure 4. Molecular interaction between beclabuvir and NS5B$_{HCV}$. Beclabuvir binds at the thumb domain (blue) of NS5B$_{HCV}$.

Figure 5. Binding site of RdRp$_{SARS-CoV-2}$ for commonly used antiviral drugs. A. Binding posture of beclabuvir at the catalytic site of RdRp$_{SARS-CoV-2}$. B. Overlapped binding sites of antiviral drugs with RdRp$_{SARS-CoV-2}$ (Molecular surface mainly in blue-gray indicates the binding site).

Table 1. Summary of molecular docking properties among antiviral drugs and RdRp$_{SARS-CoV-2}$.

FIGURE 1

A.

$\Delta G_{Vina} = - 8.0$ kcal mol^{-1}

$\Delta G_{AutoDock} = - 9.95$ kcal mol^{-1}

$\Delta G_{London} = - 9.06$ kcal mol^{-1}

$\Delta G_{GBVI / WSA} = - 6.67$ kcal mol^{-1}

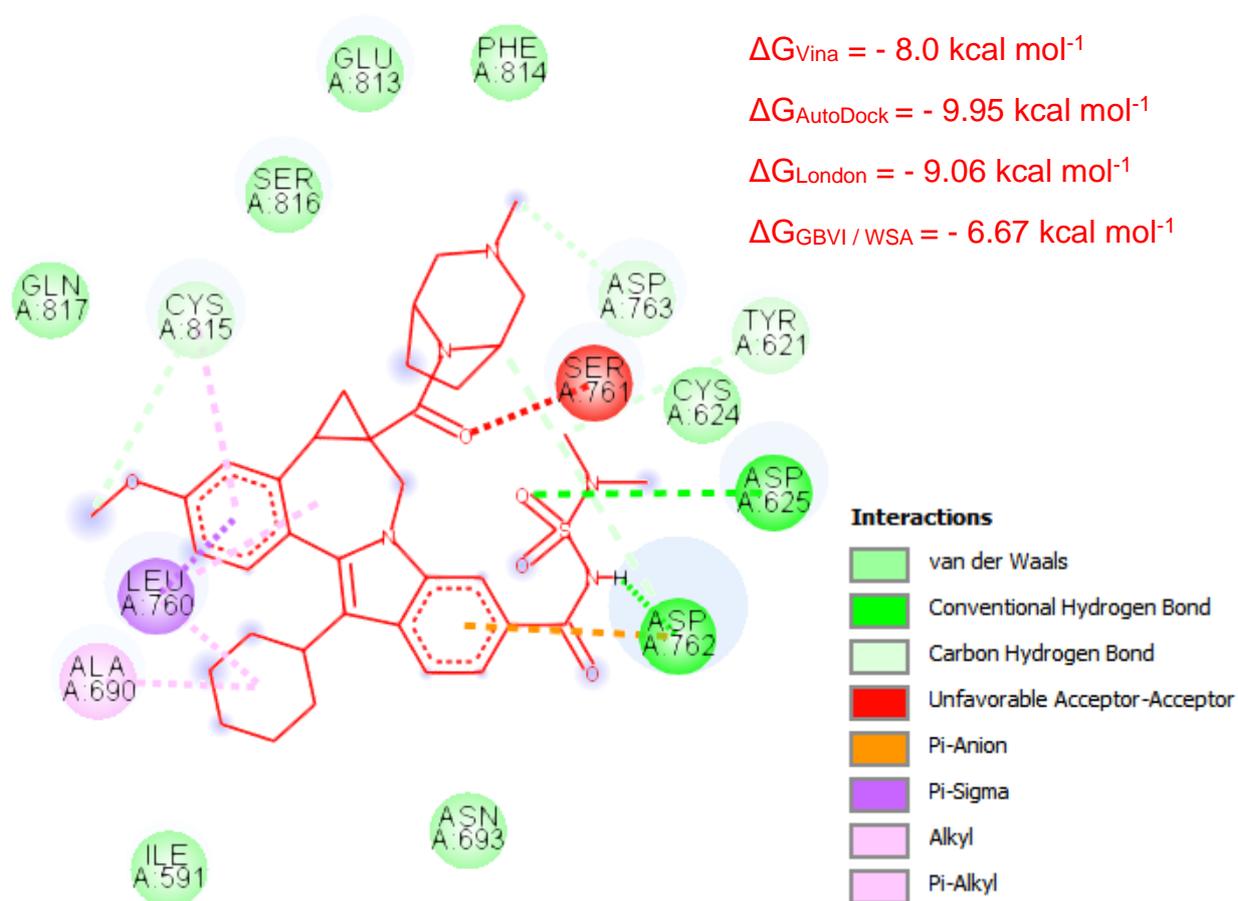



FIGURE 1

B.

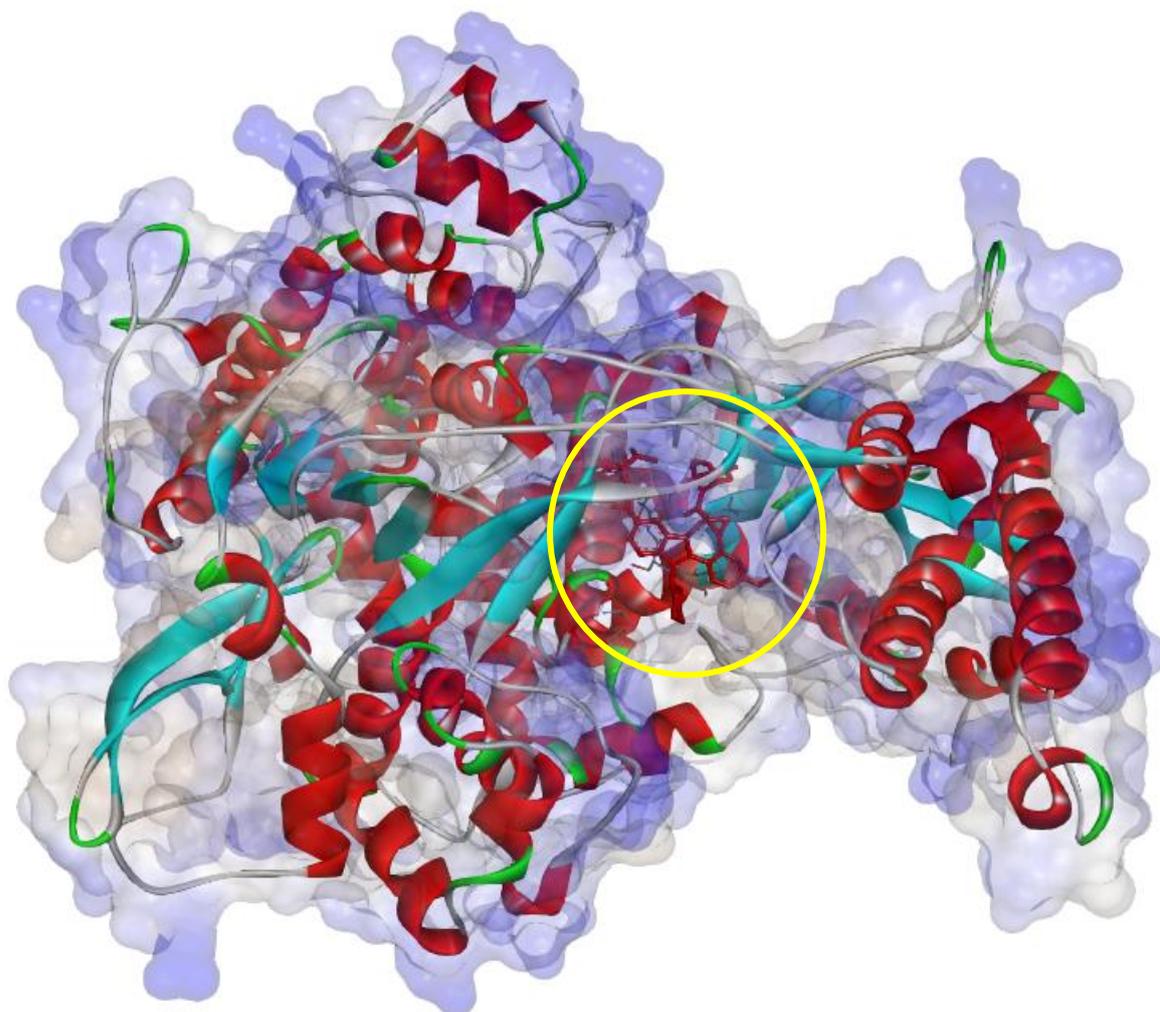



FIGURE 2

FIGURE 3

FIGURE 4

$\Delta G_{Vina} = -7.1$ kcal mol^{-1}

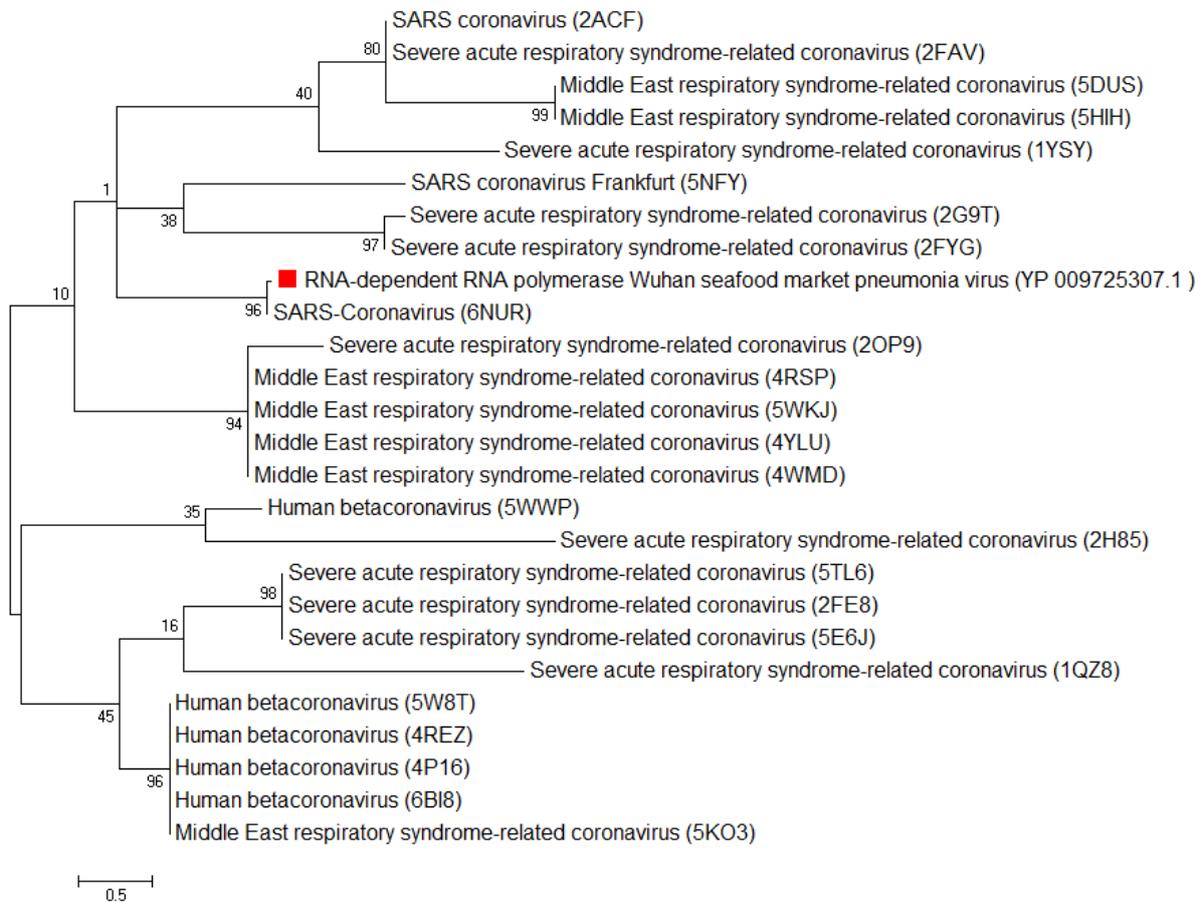
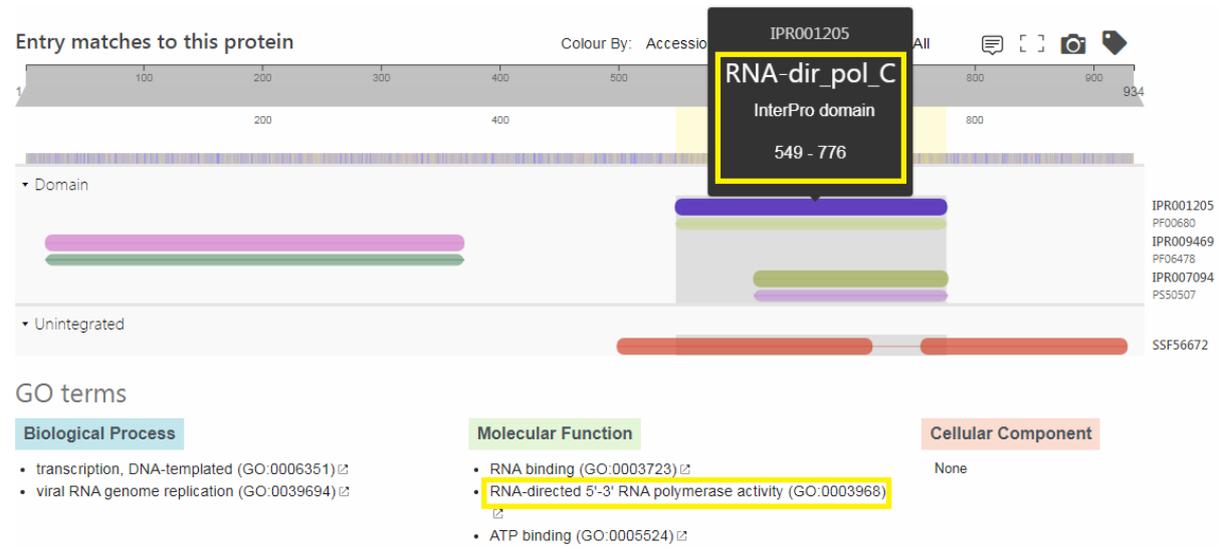
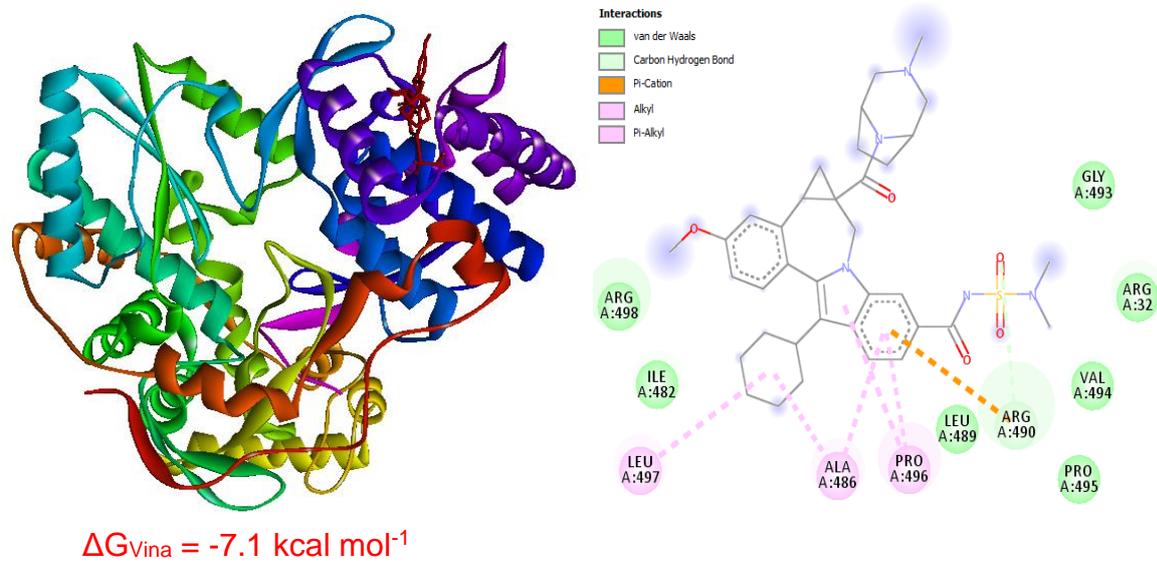
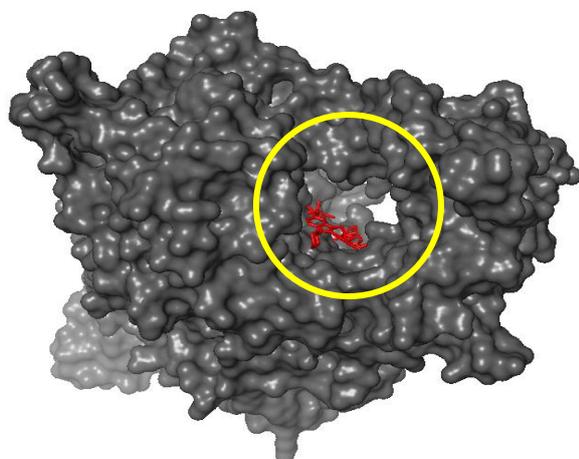
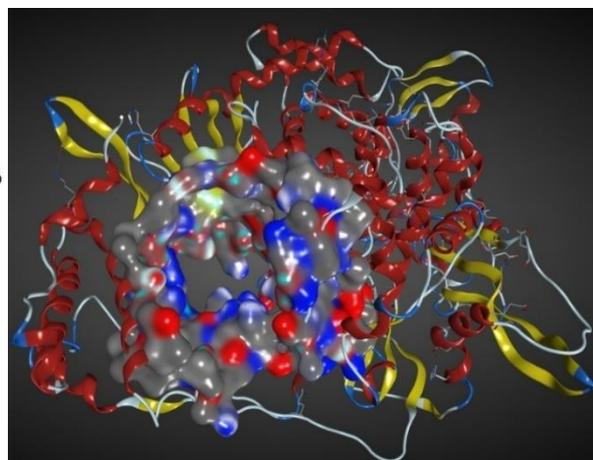



FIGURE 5

A.   **B.**

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