Investigation of anti-coronavirus, anti-HCV, nucleotide inhibitors, and bioactive molecules efficacy against RNA-directed RNA polymerase of Nipah virus: Molecular docking study

Peter T. Habib^{1*}

¹Department of Bioinformatics, Colors Medical Laboratories, Cairo, Egypt

Article Info

Article type:
Research

Article History:
Received: 2021-03-26
Accepted: 2021-05-14
Published: 2021-05-30

*** Corresponding author:**
Peter T. Habib

Department of Bioinformatics,
Colors Medical Laboratories, Cairo,
Egypt

Email: p.habib911@gmail.com

Keywords:
Molecular Docking
Nipah Virus
Antiviral Molecules and Drug

ABSTRACT

Introduction: The infections with the Nipah virus (NiV) are highly infectious and may lead to severe febrile encephalitis. High mortality rates in southeastern Asia, including Bengal, Malaysia, Papua New Guinea, Vietnam, Cambodia, Indonesia, Madagascar, the Philippines, Thailand, and India, have been reported in NiV outbreaks. Considering the high risk of an epidemic, NiV was declared a priority pathogen by the World Health Organization. However, for the treatment of this infection, there is no effective therapy or approved FDA medicines. RNA-dependent polymerase RNA (RdRp) plays an important role in viral replication among the nine well-known proteins of NiV.

Material and Methods: Fourteen antiviral molecules have been computerized for NiV RNA-dependent RNA polymerase and demonstrated a potential inhibition effect against coronavirus (NiV-RdRp). A multi-step molecular docking process, followed by extensive analyzes of molecular binding interactions, binding energy estimates, synthetic accessibility assessments, and toxicity tests.

Results: Molecular docking analysis reveals that Uprifosbuvir is the most suitable inhibitor for RdRp of Nipah Virus regarding the binding affinity and binding in the target cavity. Although, such studies need clinical confirmation.

Conclusion: The role of anti-viral molecules as a ligand against RNA-dependent RNA polymerase is critical important in the current era. Computational tools such as molecular docking has proven its power in the analysis of molecules interaction. Our analysis reveals the Uprifosbuvir might be a candidate RdRp inhibitor. This study should further investigate the properties of the already identified anti-viral molecules followed by a pharmacological investigation of these in-silico findings in suitable models.

Cite this paper as:

Habib PT. Investigation of anti-coronavirus, anti-HCV, nucleotide inhibitors, and bioactive molecules efficacy against RNA-directed RNA polymerase of Nipah virus: Molecular docking study. Front Health Inform. 2021; 10: 78. DOI: [10.30699/fhi.v10i1.288](https://doi.org/10.30699/fhi.v10i1.288)

INTRODUCTION

Nipah virus (NiV) was originally identified during an outbreak in Malaysia in 1999, was associated with the Hendra virus in the Paramyxoviridae group [1]. Since then, sporadic NiV outbreak outbreaks have been reported in several countries, including the latest in Indian Kerala [2]. NiV is endemic, highly pathogenic, and can infect most species of mammals. Henipa virus natural reservoirs seem to be Pteropus-type fruit bats [3]. However, NiV has a wide range of hosts, such as humans, leading to serious neurological, respiratory, and cardiovascular illnesses. Hosts

include horses, dogs, pigs, cats, and hamsters. Since NiV can be passed from zoonotic to human routes, the risk for outbreaks is very high [4]. Particularly, The NiV outbreaks are the most prominent for human-to-human transmission. About 600 cases of NiV human infections were reported between 1998 and 2015, where the fatality rate in Malaysia was 38% and 43-100% in India and Bangladesh [2].

Persistent pathogenicity, high mortality, and lack of effective therapeutics against NiV infection, NiV is a biosafety level 4 (BSL4) pathogen, which consider

NiV as very dangerous virus [5]. Some efforts have been made to manage NiV to search for small molecular therapy for various structural proteins [6, 7]. Favipiravir has been shown to combat viral reproduction and transcription in NiV and HeV in a recent in vitro study [8]; Also reported was R1479 (4'-azidocytidine) to inhibit various family members of Paramyxoviridae [9]. Nevertheless, no approved medicines are available for efficient human usage despite these efforts to develop such inhibitors. While Ribavirin is not an established as NiV treatment, it is especially used in emergency cases as a first-line treatment strategy for acute NiV symptoms [10]; It does, however, has different side effects during NiV therapy, such as nausea, vomiting, and convulsions [11]. Known for treating NiV, small molecules inhibit host proteins such as Interferon Regulatory Factor 3 and RIG-I-like receptors, which contribute innate immune system [12]. No approved therapeutics are available to efficiently use in humans despite these efforts to develop such inhibitors. Therefore, the definition of potential NiV inhibitors is an eminent requirement. Various efforts were made to manage NiV infection by looking for small molecular therapy for various structural proteins [6, 7, 13, 14].

The NiV virus structure is a parallel, long, coiled-coil, tetrameric with a small helical cap as tethers of the viral polymerase to nucleocapsid and multimeric phosphoprotein. Nipah virus has a single-stranded, negative-sense RNA genome encapsulated by the nucleoprotein (N) and transcribed and replicated by the polymerase protein (L). Hence molecular inhibition of RNA polymerase is one of the prime strategies for treating NiV infection. Also, as an effective strategy to interrupt virus adhesion with the host, the inferences regarding the inhibition of interactions between RNA polymerase and RNA provide the structural basis for screening antiviral therapeutic inhibitors against NiV.

The most reliable and widely used approach in the early stages of drug discovery is structure-based molecular docking [15]; however, information on their potential side effects leading to toxicity or clinical safety problems is not generally taken into account [16]. The drug's off-target toxicity and chemical structure that leads to its non-optimal efficacy [17] are predicted to cause these side effects [18]. Therefore, there is no understanding of the assessment of drug toxicity at the molecular interaction level, relying only on this method. Different methods and techniques should be used to control any possible pandemic of the Nipah virus, such as using machine learning for detection of different strains [19, 20], early detection of respiratory syndromes, and developing new drugs or scanning for the possible molecule that may target and inhibit viral life cycle [21].

Interactions between drugs and their direct target

frequently lead to differential expressions of downstream target regulated genes [22], which cause potential activation, repression, or dysregulation of downstream pathways. Here we introduce a molecular docking study for fourteen possible molecules and drug may act against RNA-dependent RNA polymerase of Nipah virus. Some previous efforts developed a vaccine based on analysis of the whole proteome [23].

MATERIAL AND METHODS

The method, software, and result analysis is imported from Habib's work [14].

Ligands retrieving and preparation

The chemical structures of selected ligands were retrieved from PubChem (<https://pubchem.ncbi.nlm.nih.gov/>), DrugBank (<https://go.drugbank.com/>), and SANCDB (<https://sancdb.rubi.ru.ac.za/>) in Protein database (PDB) format and then converted autodock ligand in the format of .pdbqt format using openbabel.

Protein Modeling, Retrieving, and Preparation

RNA-dependent RNA polymerase (RdRp) of the Nipah virus still has no identified structure. A homology model for the Nipah RdRp was built using the Swiss Model webserver [24]. We used the available RdRp sequence of the Nipah virus to build a model for the unknown structure. SWISS-MODEL shows RNA-directed RNA polymerase L of Parainfluenza as a similar structure with 36% identity and 95% coverage, which was the best result among all templates. Analysis of protein structure was quite good where Ramachandran plot archives 84.82% in the favored region (Fig 1) and MolProbity score was 1.98.

Fig 1: Ramachandran plot of RNA-dependent RNA polymerase of parainfluenza virus

Absorption, distribution, metabolism, excretion, and toxicity profile and drug ability

Pharmacokinetic parameters of a compound like absorption, distribution, metabolism, excretion, and toxicity play a vital role in the drug discovery process. Hence, an online server Swiss-Adme (<http://www.swissadme.ch/index.php>) was used to predict each ligand's physicochemical properties. We predicted GI absorption, BBB permeant, Pgp substrate, a CYP1A2 inhibitor, CYP2C19 inhibitor, CYP2C9 inhibitor, CYP2D6 inhibitor, CYP3A4 inhibitor, Lipinski violations, bioavailability score, leadlikeness, and synthetic accessibility as shown in Fig 2.

Protein-Ligand Docking

Docking was performed using autodock4 at PyRx (<https://pyrx.sourceforge.io/>) 0.8 platform. After docking, ten different poses of ligand molecules were obtained; the pose scoring lowest binding energy was chosen to visualize the ligand-protein interaction using Discovery studio 2019.

RESULTS

Swiss-Adme prediction of ligands

We studied probable physiological properties of each selected ligand to cross the blood-brain barrier, human intestinal absorptivity, caco-2 permeability, as toxicity, carcinogen character, acute oral toxicity, rat acute toxicity, and fish toxicity. Properties of selected ligand among the selected ligands, Acetaminophen were having the best lowest synthetic accessibility, which means it is easy to synthesize. Guanosine-5'-triphosphate was able to pass Blood-Brain Barrier (BBB+) and not substrate or inhibitor for different Cytochromes. On the other hand, Uprifosbuvir is intestinally absorbable while it is 545.91 in MW.

Identifying Ligand Binding Site

Ligand binding site, as the term suggests, is the site where the ligand binds. To define the binding site, we added hydrogen atom to all cavities in protein that may act as the binding site and then identify the possible binding sites from the possible receptor atoms, as it is shown in Fig 2.

Ligand Protein Docking

Setrobuvir showed the highest binding affinity with CESTP via -9.6 kcal/mol binding energy. However, the binding was not in the accurate binding site. Uprifosbuvir achieves -7.4 kcal/mol binding energy from the six binding affinity molecules, but it is still located within the binding cavity. Remdesivir is a known anti-viral drug used recently against coronavirus, which binds with -8.1 kcal/mol but

outside the ligand-binding cavity. The best molecules were Guanosine-5'-triphosphate (Fig 3), Mericitabine (Fig 4), and Uprifosbuvir (Fig 5).

Fig 2: Predicted Binding site (green) inside the protein (Gray) cavity

Fig 3: Guanosine-5'-triphosphate binding site (Left) and atoms interaction (Right)

Fig 4: Mericitabine binding site (Left) and atoms interaction (Right)

DISCUSSION

The current study was designed to understand the interaction of active molecules and drugs used recently in the coronavirus pandemic in inhibiting the Nipah virus by suppressing RNA-dependent RNA polymerase. Although X-ray crystallography and nuclear magnetic resonance techniques are utilized to understand the ligand-protein interaction, there is no known structure of RdRp.

Fig 5: Uprifosbuvir binding site (Left) and atom interaction (Right)

However, homology modeling was utilized to predict the structure of the Nipah RdRp using SWISS-MODEL. The location where the ligand should bind is identified. Molecular docking was performed on fourteen molecules and drugs to assess the binding affinity. The order of ligand according to affinity was: Setrobuvir, Remdesivir, Octahydroeuclein, Sofosbuvir, Uprifosbuvir, Adenosine-5'-triphosphate, Ribavirin, Galidesivir, Guanosine-5'-triphosphate with binding affinity -9.6, -8.1, -7.8, -7.6, -7.4, -7.4, -7, -7, -6.8 respectively. Many of them were located in the RdRp cavity, but the best affinity and target molecules were Guanosine-5'-triphosphate, Mericitabine, and Uprifosbuvir with binding affinity -6.8, -6.5, and -7.4, respectively. As shown in (Fig 5), Uprifosbuvir forms many different types of bond-like: Van der Waals, Hydrogen bond, Pi-Sigma, Amide-Pi-Stacked, Carbene-Hydrogen bond, Alkyl, and Pi-Alkyl. Guanosine-5'-triphosphate is only one form bond with an attractive charge between phosphate atom and Asparagine amino acid. After docking analysis, Uprifosbuvir was the only ligand suitable for RdRp inhibition. However, some reports show that Uprifosbuvir has a reverse or harm effect on vaccine-drug interaction. The therapeutic efficacy

of Adenovirus type 7 vaccine, Anthrax vaccine, BCG vaccine, Rubella virus vaccine, Typhoid Vaccine, and Yellow Fever Vaccine can be decreased when used in combination with Uprifosbuvir. Although, Uprifosbuvir is an approved drug for the Hepatitis C virus.

CONCLUSION

The present study represents the possible role of anti-viral molecules as a ligand against RNA-dependent RNA polymerase. Results from acute oral toxicity, rat acute toxicity, and fish toxicity predict the molecules are non-toxic, absorbable via the human intestine, and able to pass the blood-brain barrier. All the selected ligands have scored well drug-likeness scores that affect the compound's physicochemical property either towards pharmacological action or towards the drug development steps. The obtained Log S value represents the solubility of the drug molecules, which would further help in the pharmaceutical industry's formulation steps. Molecular docking study analysis reveals the Uprifosbuvir might be a candidate RdRp inhibitor. This study should encourage further investigation of the properties of the already identified anti-viral molecules followed by a pharmacological investigation of these in-silico findings in suitable models.

CONFLICTS OF INTEREST

The author declare no conflicts of interest regarding the publication of this study.

FINANCIAL DISCLOSURE

No financial interests related to the material of this manuscript have been declared.

REFERENCES

- Chua KB, Bellini WJ, Rota PA, Harcourt BH, Tamin A, Lam SK, et al. Nipah virus: A recently emergent deadly paramyxovirus. *Science*. 2000; 288(5470): 1432–5. PMID: 10827955 DOI: 10.1126/science.288.5470.1432 [[PubMed](#)]
- Chattu VK, Kumar R, Kumary S, Kajal F, David JK. Nipah virus epidemic in southern India and emphasizing “One Health” approach to ensure global health security. *J Family Med Prim Care*. 2018; 7(2): 275–83. PMID: 30090764 DOI: 10.4103/jfmpc.jfmpc_137_18 [[PubMed](#)]
- Olson JG, Rupprecht C, Rollin PE, An US, Niezgoda M, Clemins T, et al. Antibodies to Nipah-like virus in bats (*Pteropus lylei*), Cambodia. *Emerg Infect Dis*. 2002; 8(9): 987–8. PMID: 12194780 DOI: 10.3201/eid0809.010515 [[PubMed](#)]
- Luby SP. The pandemic potential of Nipah virus. *Antiviral Res*. 2013; 100(1): 38–43. PMID: 23911335 DOI: 10.1016/j.antiviral.2013.07.011 [[PubMed](#)]
- Freiberg AN, Worthy MN, Lee B, Holbrook MR. Combined chloroquine and ribavirin treatment does not prevent death in a hamster model of Nipah and Hendra virus infection. *J Gen Virol*. 2010; 91(Pt 3): 765–72. PMID: 19889926 DOI: 10.1099/vir.0.017269-0 [[PubMed](#)]
- Tigabu B, Rasmussen L, White EL, Tower N, Saeed M, Bukreyev A, et al. A BSL-4 high-throughput screen identifies sulfonamide inhibitors of Nipah virus. *Assay Drug Dev Technol*. 2014; 12(3): 155–61. PMID: 24735442 DOI: 10.1089/adt.2013.567 [[PubMed](#)]
- Niedermeier S, Singethan K, Rohrer SG, Matz M, Kossner M, Diederich S, et al. A small-molecule inhibitor of Nipah virus envelope protein-mediated membrane fusion. *J Med Chem*. 2009; 52(14): 4257–65. PMID: 19499921 DOI: 10.1021/jm900411s [[PubMed](#)]

8. Dawes BE, Kalveram B, Ikegami T, Juelich T, Smith JK, Zhang L, et al. Favipiravir (T-705) protects against Nipah virus infection in the hamster model. *Sci Rep.* 2018; 8(1): 7604. PMID: 29765101 DOI: 10.1038/s41598-018-25780-3 [[PubMed](#)]
9. Hotard AL, He B, Nichol ST, Spiropoulou CF, Lo MK. 4'-Azidocytidine (R1479) inhibits henipaviruses and other paramyxoviruses with high potency. *Antiviral Res.* 2017; 144: 147–52. PMID: 28629988 DOI: 10.1016/j.antiviral.2017.06.011 [[PubMed](#)]
10. Snell NJC. Ribavirin therapy for Nipah virus infection. *J Virol.* 2004; 78(18): 10211. PMID: 15331756 DOI: 10.1128/JVI.78.18.10211.2004 [[PubMed](#)]
11. Chong H-T, Kamarulzaman A, Tan C-T, Goh K-J, Thayaparan T, Kunjapan SR, et al. Treatment of acute Nipah encephalitis with ribavirin. *Ann Neurol.* 2001; 49(6): 810-3. PMID: 11409437 DOI: 10.1002/ana.1062 [[PubMed](#)]
12. Guillaume V, Contamin H, Loth P, Grosjean I, Courbot MCG, Deubel V, et al. Antibody prophylaxis and therapy against Nipah virus infection in hamsters. *J Virol.* 2006; 80(4): 1972–8. PMID: 16439553 DOI: 10.1128/JVI.80.4.1972-1978.2006 [[PubMed](#)]
13. Habib PT. Vaccine design, adaptation, and cloning design for multiple epitope-based vaccine derived from SARS-CoV-2 surface glycoprotein (S), membrane protein (M) and envelope protein (E): In silico approach. *Front Health Inform.* 2021; 10: 67.
14. Habib PT, Saber-Ayad M, Hassanein SE. In silico analysis of 716 natural bioactive molecules from Atlantic ocean reveals candidate molecule to inhibit spike protein [Internet]. 2021 [cited: 2021 Mar 3]. Available from: <https://www.researchsquare.com/article/rs-156623/v1>
15. Meng X-Y, Zhang H-X, Mezei M, Cui M. Molecular docking: A powerful approach for structure-based drug discovery. *Curr Comput Aided Drug Des.* 2011; 7(2): 146-57. PMID: 21534921 DOI: 10.2174/157340911795677602 [[PubMed](#)]
16. Kola I, Landis J. Can the pharmaceutical industry reduce attrition rates? *Nat Rev Drug Discov.* 2004; 3(8): 711-5. PMID: 15286737 DOI: 10.1038/nrd1470 [[PubMed](#)]
17. Bender A, Scheiber J, Glick M, Davies JW, Azzaoui K, Hamon J, et al. Analysis of pharmacology data and the prediction of adverse drug reactions and off-target effects from chemical structure. *Chem MedChem.* 2007; 2(6): 861-73. PMID: 17477341 DOI: 10.1002/cmdc.200700026 [[PubMed](#)]
18. Brouwers L, Iskar M, Zeller G, van Noort V, Bork P. Network neighbors of drug targets contribute to drug side-effect similarity. *PloS One.* 2011; 6(7): e22187. PMID: 21765950 DOI: 10.1371/journal.pone.0022187 [[PubMed](#)]
19. Habib PT, Alsamman AM, Saber-Ayad M, Hassanein SE, Hamwieh A. COVIDier: A deep-learning tool for coronaviruses genome and virulence proteins classification [Internet]. 2020 [cited: 2021 Mar 3]. Available from: <https://www.biorxiv.org/content/10.1101/2020.05.03.075549v1.abstract>
20. Habib PT. COVATOR: A Software for chimeric coronavirus identification [Internet]. 2020 [cited: 2021 Mar 3]. Available from: <https://www.biorxiv.org/content/10.1101/2020.11.14.383075v1.abstract>
21. Habib PT, Alsamman AM, Hassanein SE, Hamwieh A. Developing convolutional neural networks-based system for predicting pneumonia using X-radiography image. *Highlights in BioScience.* 2020; 3: 1-3.
22. Noh H, Shoemaker JE, Gunawan R. Network perturbation analysis of gene transcriptional profiles reveals protein targets and mechanism of action of drugs and influenza: A viral infection. *Nucleic Acids Res.* 2018; 46(6): e34. PMID: 29325153 DOI: 10.1093/nar/gkx1314 [[PubMed](#)]
23. Habib PT. Learning from COVID-19 pandemic: In silico vaccine and cloning design against Nipah virus by studying and analyzing the whole Nipah virus proteome [Internet]. 2021 [cited: 2021 Mar 3]. Available from: <https://www.researchsquare.com/article/rs-269666/v1>
24. Biasini M, Bienert S, Waterhouse A, Arnold K, Studer G, Schmidt T, et al. SWISS-MODEL: Modelling protein tertiary and quaternary structure using evolutionary information. *Nucleic Acids Res.* 2014; 42(Web Server Issue): W252-8. PMID: 24782522 DOI: 10.1093/nar/gku340 [[PubMed](#)]