Study of Novel Potential Antiviral Drug Candidates Using Computational Tools

Summary

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Chapter 1: Introduction

A virus is a microscopic infectious agent that can only replicate inside the living cells of another organism. They are composed of a protein coat surrounding genetic material, which can be either DNA or RNA. Viral diseases emerge as the leading causes of human and animal mortality, contributing to high healthcare expenditure around the World. There are many types of viruses, each with its own unique characteristics and targets. Some common viruses are Influenza virus, HIV, Coronavirus, Hepatitis virus, etc. The mankind has seen the outbreaks and epidemics of various chronic diseases (Ebola virus, Zika virus, influenza virus, SARS, MERS, etc.). In the past, however, significant progress was made in combating major epidemic diseases such as COVID-19, influenza, and Ebola.

To battle these viruses, antiviral drugs are frequently used to inhibit the replication and spread of viruses within the body. Although there are several antiviral drugs already available in the market to treat different viral diseases, but still there is always need of modification and designing of novel drugs in order to increase their efficacy. Antiviral drug discovery is a critical area of research due to the emergence of new viral infections and the development of drug resistance. The conventional method of antiviral drug designing requires skilled labours, high technology equipment, budget of $1.8 to $2 billion and an average of 15 to 20 years.

To cut-short the limitations of conventional method, computational simulations in the form of Computer-Aided Drug Designing (CADD) are employed as a modern technique of drug designing. The method explores vast chemical spaces, predict molecular interactions, and optimize drug properties with precision. It utilizes molecular modelling tools to predict the interactions between drug candidates and biological targets. Various molecular modelling techniques are employed in CADD, including docking and molecular dynamics, which are necessary for evaluating the binding affinities and interactions of compounds. It facilitates the rational design of drug candidates, enhancing efficacy while reducing harmful side effects. By leveraging computational algorithms and simulations, it also helps in identifying promising lead

compounds, prioritize them for synthesis and biological testing, and optimize their chemical structures. With the increasing complexity of diseases and the need for targeted therapies, CADD plays an important role in identifying innovative drug candidates and addressing unmet medical needs effectively. **Figure 1** illustrates the role of CADD in identifying drug candidates. Hence, by employing the methods of CADD the drug designing process can be made faster, easier, and more effective.

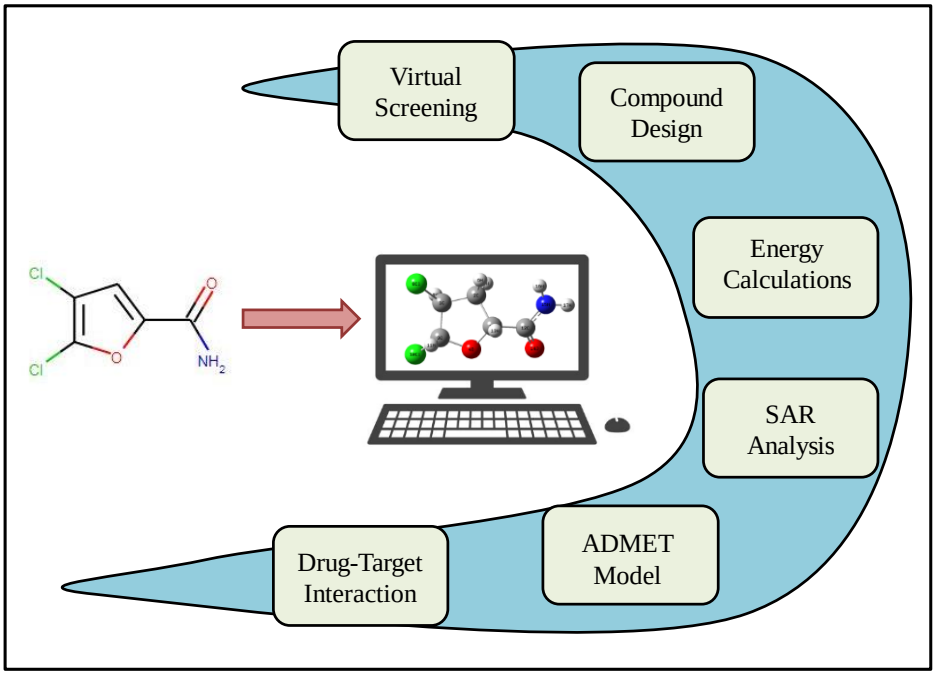


Figure 1: Role of CADD in identifying drug candidates.

The aim of proposed research is to design novel potential antiviral drug candidates by employing computational methods and tools such as fragment-based drug designing, optimization, pharmacokinetics, Molecular Docking, Molecular Dynamics (MD) and MM/PBSA. A thorough study of physical and biological properties, and interactions would provide a theoretical protocol for designing new antiviral drugs.

Chapter 2: Methodology

Main methods used in the present research are discussed below:

Fragment-Based Drug Designing

Fragment-Based Drug Design (FBDD) focuses on the identification and optimization of small molecular fragments that bind weakly to target proteins. Lead compounds designed via FBDD have higher success rate since it places an emphasis on efficiency and design. FBDD offers several advantages over traditional drug discovery

methods, including a reduced risk of false positives, increased chemical diversity, and the potential for discovering novel binding modes. To identify the fragments, U.S. FDA approved antiviral drugs are considered, where core is identified and moieties are grown around it resulting in a new compound or scaffold. The screening of the compounds is usually done on the basis of ligand efficacy and drug-likeness.

Density Functional Theory

Density Functional Theory (DFT) is a computational quantum mechanical modelling method used to study the electronic structure (ideally the ground state) of many-body system, molecules and the condensed phases. The properties of the system are studied by using functional, i.e. mapping of one function onto another function, which in this case is the spatially dependent electron density. DFT is a popular and versatile method in field of biophysics, computational physics and computational chemistry. The geometry optimization and structural properties of the designed compounds were achieved using Gaussian 09, GAMESS and Quantum Espresso software packages.

Molecular Docking

Molecular docking is a computational technique used to predict the binding orientation and affinity of a molecule (ligand) to a target protein. It involves creating a three-dimensional structure of the protein and ligand, and then using algorithms to search for the most energetically favourable binding conformation. This involves considering factors such as electrostatic interactions, hydrogen bonding, van der Waals forces, and hydrophobic interactions. Molecular docking is a valuable tool in drug discovery, as it can help identify potential drug candidates by predicting their binding affinity to a target protein. Autodock 4.2 software was used to perform molecular docking and study the interactions of designed compounds with the target. AutoDock employs Lamarkian Genetic Algorithm (LGA) to search for the optimal binding pose of a ligand to a protein. It can handle both flexible ligands and flexible proteins, making it a versatile tool for drug discovery.

Molecular Dynamics Simulation

Molecular dynamics (MD) simulation is a computational method used to study the dynamics and behaviour of molecules at an atomic or molecular level. It involves

applying classical mechanics principles to simulate the motion of atoms and molecules over time. By calculating the forces between atoms and integrating Newton's equations of motion, molecular dynamics simulations can provide insights into a wide range of phenomena, including drug-protein interactions, protein folding, enzyme catalysis, and materials properties. In MD simulation, the atoms and molecules are allowed to interact for a period of time, such that the motion of the atoms can be studied and the obtained results are analysed in the form of trajectories. The complex is put in a water box and relaxed by a series of constrained energy minimization and Molecular Mechanics (MM) runs at MM level. GROMACS and YASARA were used to perform molecular dynamics of the selected docked poses.

Molecular Mechanics/Poisson-Boltzmann Surface Area (MM/PBSA)

MM/PBSA is a computational method used to calculate the binding free energy of a ligand to a protein. It combines two main components "Molecular Mechanics (MM)" and "Poisson-Boltzmann (PB) Surface Area (SA)" The MM involves using classical mechanics to calculate the potential energy of the system, including bond stretching, angle bending, torsions, and non-bonded interactions (van der Waals and electrostatic). Whereas, the PB solves the Poisson-Boltzmann equation to calculate the electrostatic solvation free energy of the system. It considers the interactions between the protein and ligand with the surrounding solvent. The SA calculates the solvent-accessible surface area of the protein and ligand. This term is used to estimate the non-polar solvation free energy based on the hydrophobic effect. MM/PBSA provides a relatively fast and accurate estimate of binding free energy between a protein and ligand, aiding in the identification and optimization of potential drug candidates.

Pharmacokinetics

Pharmacokinetics (PK) is the branch of pharmacology dedicated to the study of how drugs move through the body over time. It involves the analysis of the absorption, distribution, metabolism, excretion and toxicity (ADMET) of drugs. The primary goal of pharmacokinetics is to understand the drug concentration at the site of action and its variation with time. The foundation of pharmacokinetics is mathematical modelling, which emphasizes the correlation between drug plasma levels and time since administration. The pharmacokinetics study was performed using SwissADME and admetSAR webservers.

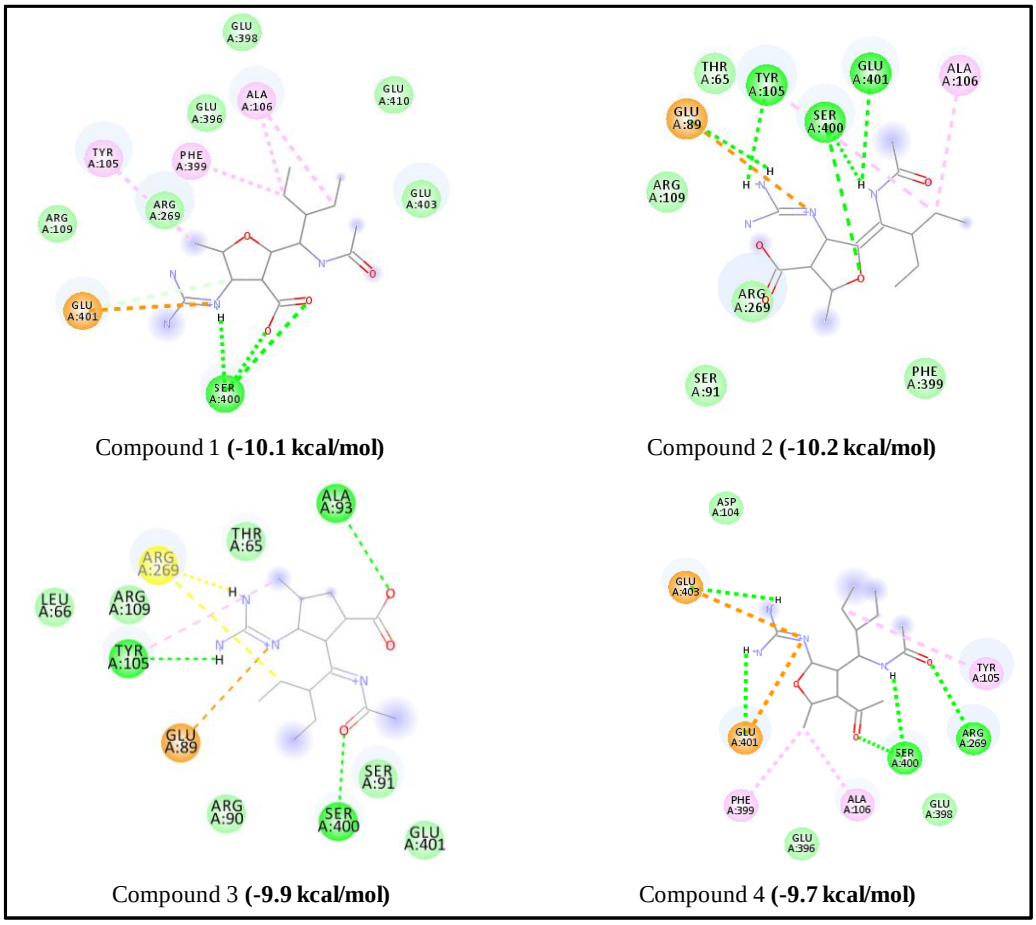
Chapter 3: Derivatives of 2-methyl tetrahydrofuran-n-carboxylic acids as Potential Inhibitors Against Hemagglutinin HA3 of Influenza A Virus

Chapter 3 focuses on developing inhibitors for the novel HA3 subtype of Influenza A Virus Hemagglutinin, which has pandemic potential as reported by the WHO. The study involves generating a phylogenetic tree to understand the sequence similarity among 18 hemagglutinin subtypes and comparing it to H3N2. Four compounds were selected from scaffold structures based on FDA-approved Peramivir for their drug-likeness and synthetic feasibility. These compounds underwent theoretical evaluations, including electronic, spectroscopic, ADMET analysis, molecular docking and molecular dynamic to validate their potential as drug candidates.

The study identified that derivatives of 2-methyl tetrahydrofuran-n-carboxylic acid designed using fragment-based drug designing of Peramivir, exhibit inhibitory activity against H3N2 with stable conformations. The research methodology included phylogenetic analysis of influenza A hemagglutinin genomes, geometrical optimization of the designed scaffolds, spectroscopic analysis, physicochemical properties evaluation, pharmacokinetics analysis, molecular docking, and molecular dynamics simulations.

The virtually designed drugs were studied under geometrical and spectroscopic conditions, it turned out that the designed molecules exhibited stable conformation in terms of energy and structural feasibility. Peramivir exhibited lower zero-point energy as compared to the virtually designed drugs which clearly indicated that the designed compounds possess stable energy in quantum mechanical state. Since, Peramivir is FDA approved and the reported compounds are its scaffold structure therefore the ADMET parameters are fulfilled, but to gain further insight pharmacokinetic analysis has been done to validate the biochemical activity. The screened compounds were subjected to molecular docking. The strong binding affinities were obtained for compound 1, compound 2, compound 3 and compound 4 to be -10.1 kcal/mol, -10.2 kcal/mol, -9.9 kcal/mol and -9.7 kcal/mol, respectively. These designed compounds show better binding affinity, than FDA approved Peramivir with H7N9 (-7.2 kcal/mol) as reported by *Su et al. (2013)*. All the four reported drug compounds perfectly bind to the cavity and forms stable bonds with Ser400 and Tyr105. The reported compounds

made hydrogen bonds with Ser400, Glu401, Glu403, Tyr105, Thr65, Ala93 and Arg269 in different conformations, thereby showing the stability of compounds in the binding region. **Figure 2** shows the docked complexes of designed compounds.



Compound 1 **(-10.1 kcal/mol)** Compound 2 **(-10.2 kcal/mol)**

Compound 3 **(-9.9 kcal/mol)** Compound 4 **(-9.7 kcal/mol)**

Figure 2: 2D representation of docked complexes with PDB ID: 4WEA.

To validate the docking results MD was performed. The stable MD simulation validates the conformational stability and flexibility of the potent hit compounds within the binding cavity. Root Mean Square Deviation (RMSD) of the reported complexes were studied, revealing that the stability was achieved after 3 ns for all 4 docked complexes. The RMSD values were observed to be within the range 1.5-2.23 Å after beginning from 0.5 Å, as illustrated in **Figure 3**. Similar deviation pattern was observed for compound 1, compound 4 and Peramivir at every 20 ns time steps, this signifies that these molecules are undergoing regular conformational changes and oscillations. This is due to the factors such as internal vibrations and interactions with the solvent molecules. Therefore, the properties of compound 1 and compound 4 are closely related to that of Peramivir.

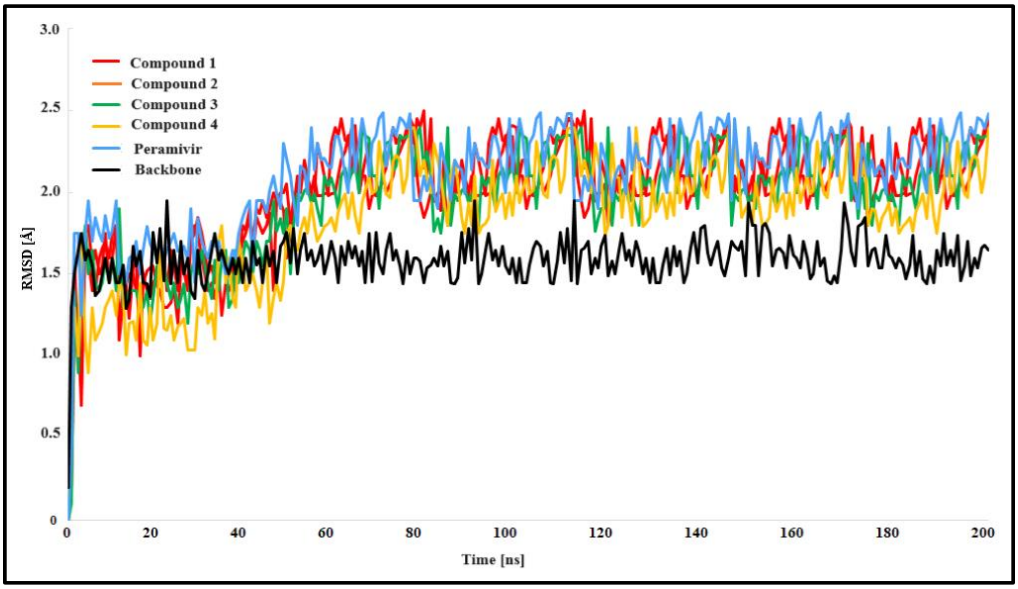


Figure 3: RMSD plot of docked complexes.

The phylogenetic analysis revealed that the HA3 subtype shares significant genomic similarity with other influenza subtypes, suggesting that the inhibitors designed could potentially be effective against similar variants. Therefore, the four reported compounds could be promising candidates for further laboratory synthesis and clinical trials, with the potential to combat various influenza variants.

Chapter 4: Exploration of Selective Inhibitor Against Variola Virus Thymidylate Kinase

Chapter 4 focuses on the development of a potential inhibitor for thymidylate kinase (TMPK) from the Variola virus, closely related to the monkeypox virus. Given the resurgence of monkeypox cases, the study aims to design an alternative to Tecovirimat (TPOXX), the only FDA-approved antiviral drug for treating Orthopoxviruses like monkeypox and smallpox. The study deals with the application of Fragment-Based Drug Design (FBDD) approach to develop a new compound, POX-A, and carried out a detailed computational analysis to compare its efficacy against TPOXX. The analysis includes molecular docking, dynamics simulations, and binding free energy calculations.

The study uses the protein structure of Vaccinia virus TMPK as a template to model the Variola virus TMPK (VrTMPK) and conducts docking studies to assess the interaction between POX-A and both VrTMPK and human TMPK (HssTMPK). Further, using FBDD to generate POX-A based on the structure of TPOXX. The goal

was to create a more stable and selective inhibitor by altering the chemical structure of TPOXX. Computational methods such as Density Functional Theory (DFT) were applied to determine the structural and electronic properties of POX-A. The results indicated that POX-A had superior stability and electronic characteristics compared to TPOXX. A pharmacokinetic analysis also revealed that POX-A met the necessary criteria for drug-likeness, including favourable ADMET properties.

The reported compound was used in the docking simulation with the binding cavities of VrTMPK and HssTMPK, which is in the form of a complex with thymidine 5'-diphosphate (TDP) and cofactor Mg^{2+}. The results of docking studies predicted interaction energy ($E_{interaction}$), interaction energy of the cofactor ($E_{cofactor}$), H-bond energy, residues interacting to form H-bond, and the difference of interaction energy of VrTMPK and HssTMPK ($\Delta E^{i}_{interaction}$; where i is the difference of interaction energy of VrTMPK and HssTMPK) are given in **Table 1**. The results of the docking studies show that POX-A has a higher affinity for VrTMPK than for HssTMPK, indicating its potential as a selective inhibitor for the viral enzyme.

Table 1: Docking results of POX-A and TPOXX with VrTMPK and HssTMPK.

Compound	$E_{interaction}$		$E_{cofactor}$		Hydrogen Bond Energy		Hydrogen Bond Interaction with Residues		$\Delta E^{i}_{interaction}$
	HssTMPK	VrTMPK	HssTMPK	VrTMPK	HssTMPK	VrTMPK	HssTMPK	VrTMPK	
POX-A	-73.55	-135.46	-0.22	-5.78	-5.24	-9.33	Arg76 Arg97	Arg41 Lys17 Arg93 Phe38 Arg 72	61.91
TPOXX	-70.31	-130.71	-0.19	-4.37	-4.96	-8.77	Arg97	Lys17 Arg41 Arg93	53.99

Molecular dynamics (MD) simulations of the docked complexes over 200 nanoseconds confirm the stability of POX-A when bound to VrTMPK, with the complex exhibiting a lower Root Mean Square Deviation (RMSD) than the complex with HssTMPK. The H-bond analysis confirmed the formation of hydrogen bonds in and around the active site of VrTMPK. Therefore, the results of MD simulation were in

good agreement to the results obtained from docking studies. This signifies the higher stability of the complex.

MM/PBSA calculations were performed to understand the binding free energy constituents and stability of the POX-A-VrTMPK complex. The results indicate that POX-A has a higher binding free energy with VrTMPK than with HssTMPK, further supporting its potential as a selective inhibitor. **Table 2** shows the results of MM/PBSA analysis. VrTMPK exhibited the binding free energy -163±26.30 kJ/mol for POX-A which was calculated by MM/PBSA method. These results of POX-A were compared with TPOXX.

Table 2: Tabular representation for the results of MM/PBPSA.

MM/PBPSA Parameters (kJ/mol)	**HssTMPK**	**VrTMPK**
$\boldsymbol{\Delta E_{vdW}}$	-222.76±20.44	-240.63±23.74
$\boldsymbol{\Delta E_{ele}}$	-78.18±12.46	-51.74±9.11
$\boldsymbol{\Delta G_{pol}}$	245.27±51.42	150.33±6.87
$\boldsymbol{SASA}$	-20.04±43.33	-21.41±0.32
$\boldsymbol{\Delta G_{bind}}$	**-75.71±24.81**	**-163.45±26.30**

The study presents POX-A as a promising alternative to TPOXX for the inhibition of thymidylate kinase and a selective inhibitor for VrTMPK, thus possibly a novel drug compound for Monkeypox and Variola Virus. The computational analysis showed that POX-A is more stable and has a stronger binding affinity for the viral TMPK, making it a potential candidate for further in vitro and in vivo testing. This study highlights the importance of computational approaches in the early stages of drug development and paves the way for the design of more effective antiviral therapies.

Chapter 5: Molecular Docking and Molecular Dynamics Based Study for Identification of Ebola Inhibitors

Chapter 4 focuses on the design and analysis of potential antiviral compounds targeting Ebola virus receptors using computer-aided drug design (CADD) methods. The study begins by emphasizing the need for new drugs to treat Ebola, a deadly virus, by identifying inhibitors for key viral proteins like VP24, VP30, VP35, and VP40. Galidesivir (BCX4430) is a known antiviral but the study aims to enhance its molecular

structure. The study is focused on designing eight pyrrolopyrimidine derivatives and evaluated their antiviral activity using various computational techniques including Density Functional Theory (DFT) and Molecular Dynamics (MD) simulations.

The pharmacokinetic analysis of the eight compounds revealed four compounds as promising candidates. These were evaluated based on their drug-likeness and ADMET properties. Compound 4 stood out, demonstrating favourable pharmacokinetics and physiochemical characteristics. Following this, DFT calculations were performed to understand electronic properties like the HOMO-LUMO energy gap, which provides insight into chemical reactivity and stability.

Screened compound 4 was subjected to molecular docking. Compound 4 exhibited higher stability as compared to BCX4430, a critical factor for drug effectiveness. **Figure 4** illustrates the docked complexes of compound 4 with the four receptors of Ebola virus. The minimum binding energy recorded for the interaction of compound 4 was -9.8 kcal/mol, -9.6 kcal/mol, -9.7 kcal/mol, -9.5 kcal/mol for VP24, VP30, VP35, and VP40, respectively.

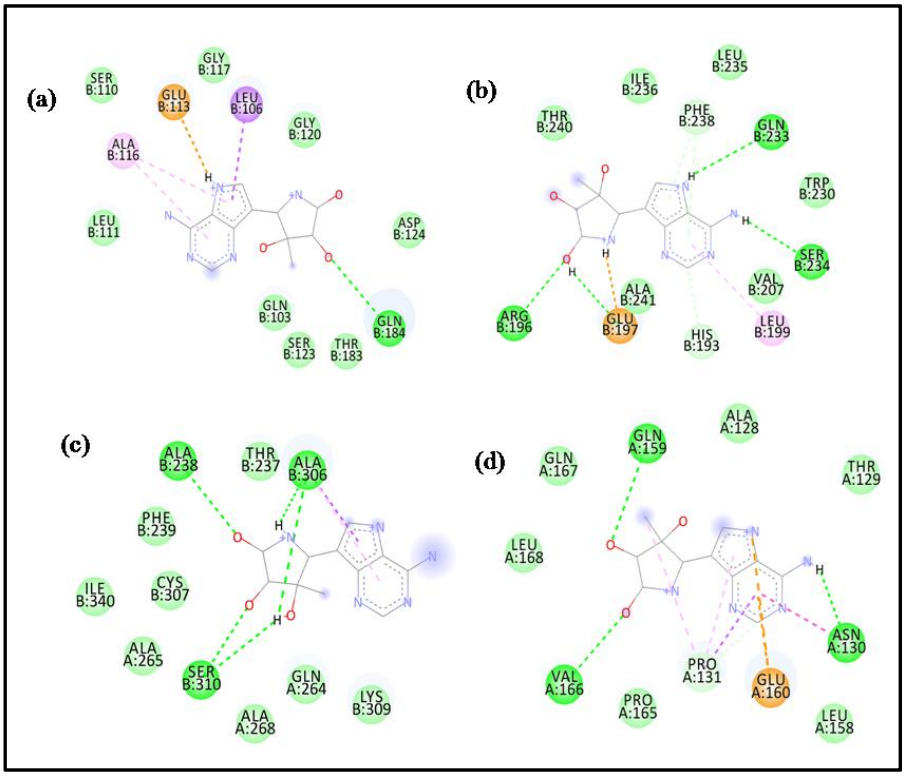


Figure 4: 2D interaction of compound 4 with VP24, VP30, VP35 and VP40, respectively.

The study highlighted hydrogen bonds and hydrophobic interactions between the compound and receptors, further validating its potential as a drug candidate. The docking analysis confirmed that Compound 4 made better interactions with the viral receptors, surpassing the BCX4430 in binding efficiency.

The stability of the docked complexes was then analysed using MD simulations over 100 nanoseconds. Root Mean Square Deviation (RMSD), Root Mean Square Fluctuation (RMSF), and Radius of Gyration (Rg) were used to assess the dynamic behaviour of the receptor-ligand complexes. Compound 4 demonstrated greater stability across the viral proteins than BCX4430. Additionally, MM/PBSA method was employed to calculate the binding free energies, which further reinforced Compound 4 has higher binding free energy to Ebola receptors. **Table 3** shows the computed MM/PBSA parameters.

Table 3: Computed MM/PBSA parameters.

Parameters (kcal/mol)	**VP24**	**VP30**	**VP35**	**VP40**
		Compound 4		
$\boldsymbol{\Delta E_{vdW}}$	-223.45±21.32	-248.19±22.61	-201.36±26.79	-219.88±29.94
$\boldsymbol{\Delta E_{ele}}$	-75.12±11.20	-52.34±12.53	-49.18±12.36	-53.49±11.09
$\boldsymbol{\Delta G_{pol}}$	146.23±61.21	201.79±59.64	105.34±15.29	106.49±17.44
$\boldsymbol{SASA}$	-15.3±44.20	-39.45±45.20	-25.39±22.36	-41.08±21.83
$\boldsymbol{\Delta G_{bind}}$	**-167.64±15.51**	**-138.28±21.00**	**-170.59±46.22**	**-207.96±39.42**
		BCX4430		
$\boldsymbol{\Delta E_{vdW}}$	-150.37±10.25	-152.33±12.51	-195.35±12.34	-183.48±15.37
$\boldsymbol{\Delta E_{el}}$	-21.38±7.51	-21.24±6.38	-75.40±7.38	-62.28±8.67
$\boldsymbol{\Delta G_{pol}}$	25.99±9.50	75.63±17.26	192.55±9.08	117.66±9.27
$\boldsymbol{SASA}$	-7.81±10.34	-5.40±10.22	-13.25±11.67	-21.45±12.41
$\boldsymbol{\Delta G_{bind}}$	**-153.57**±18.60	**-103.38±11.85**	**-91.45±22.31**	**-149.55±27.18**

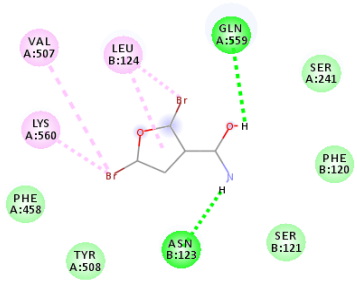
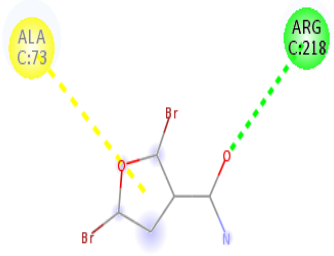
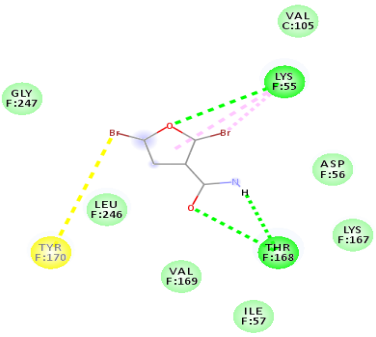
Therefore, the study reveals Compound 4 as a strong drug candidate for treating Ebola. The combination of virtual screening, DFT analysis, docking studies, and MD simulations provides compelling evidence that this compound outperforms BCX4430 in binding and stability with key Ebola virus receptors. Hence, the study lays important groundwork for future antiviral drug development against Ebola.

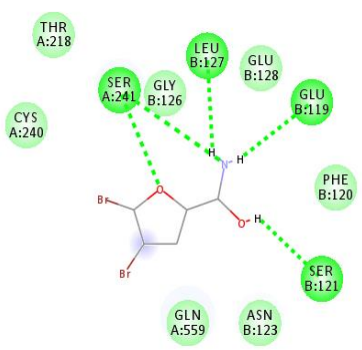
Chapter 6: Identification of Novel Potential Multitarget Inhibitors for Nipah Virus by *in silico* Route

Chapter 6 discusses the design of multitarget antiviral inhibitors against the Nipah virus using computational approaches. Nipah virus, a deadly pathogen causing encephalitis and respiratory illness, has no specific antiviral treatment. In the study, twelve potential drug compounds were designed based on Favipiravir, an antiviral known for its activity against RNA viruses. Amongst twelve, five compounds exhibited strong drug-likeness and favourable ADMET properties, making them viable candidates for further analysis. These compounds were evaluated using molecular docking, molecular dynamics simulations, and binding energy calculations to assess their inhibitory potential against three key Nipah virus proteins: NiV-G, NiV-F, and NiV-N.

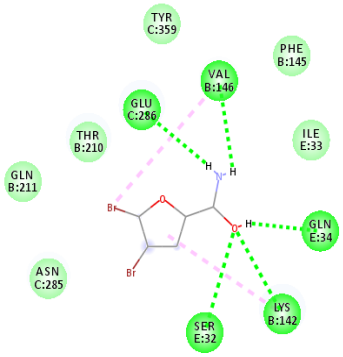
The study utilized FBDD to generate derivatives of Favipiravir, and molecular docking studies were conducted to evaluate how well these compounds bind to the viral proteins. Compounds C3, C4, C5, C6, and C7 showed the most promising binding affinities, indicating their potential as inhibitors. **Table 4** tabulates the 2D interactions of best docked poses.

Table 4: 2D interactions of best docked poses (green, yellow and pink residues with bond represents H-bond, hydrophobic and alkyl interactions).

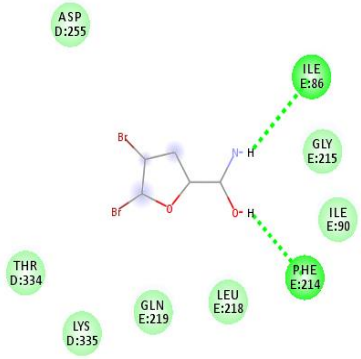
		
C3 with NiV-G	**C3 with NiV-F**	**C3 with NiV-N**



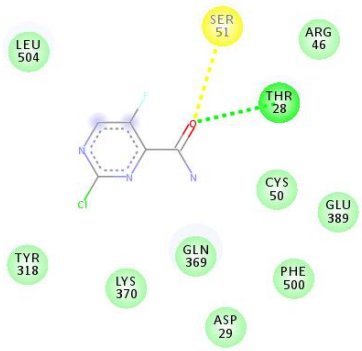
C4 with NiV-G



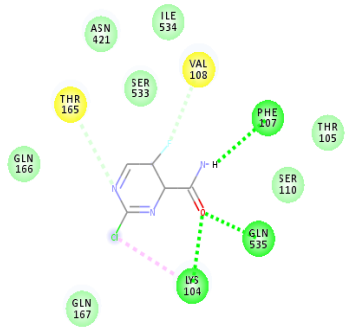
C4 with NiV-F



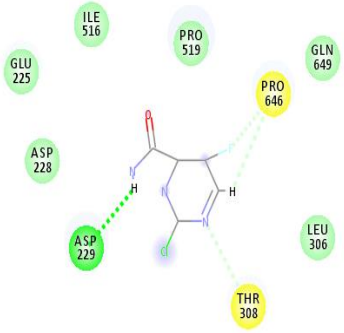
C4 with NiV-N



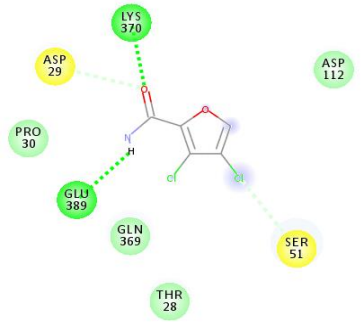
C5 with NiV-G



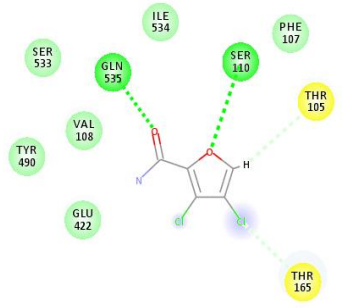
C5 with NiV-F



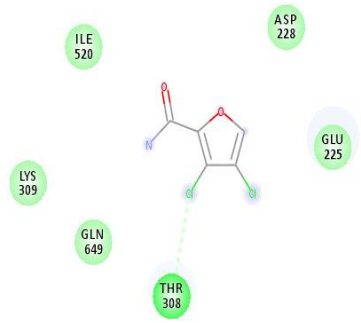
C5 with NiV-N



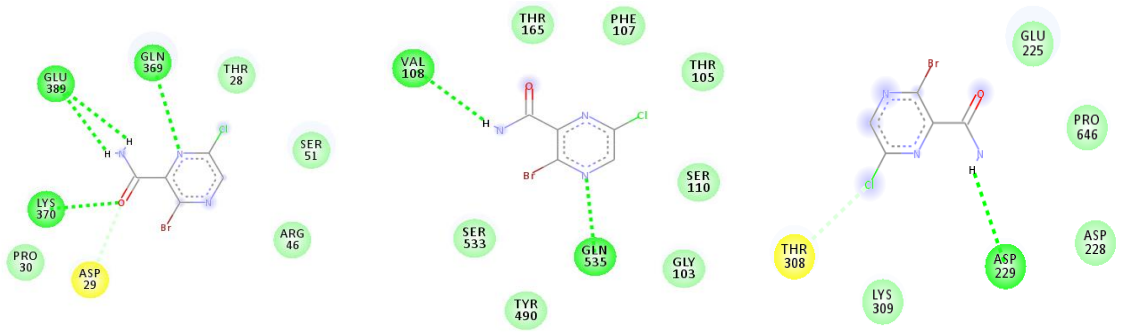
C6 with NiV-G



C6 with NiV-F



C6 with NiV-N



C7 with NiV-G **C7 with NiV-F** **C7 with NiV-N**

Molecular dynamics (MD) simulations confirmed the stability of these compounds when bound to the target proteins, with Root Mean Square Deviation (RMSD) values indicating minimal structural fluctuations, which is a key indicator of effective binding. The MD results were in good agreement with the docking simulations. The MM/PBSA analysis was also performed to calculate the binding free energies of the docked compounds. This analysis provided further evidence of the strong interactions of compound with the viral proteins. Compounds C4, C6, and C7, in particular, exhibited higher binding free energies, indicating their superior ability to bind to and inhibit the virus's key proteins. **Table 5** represents the parameters of binding free energy. These findings suggest that these compounds could serve as effective multitarget inhibitors of Nipah virus.

Table 5: Parameters of binding free energy for Favipiravir derivatives with multitarget of NiV (Energies are calculated in kcal/mol).

Compounds	**NiV-G**				
	ΔE_{ele}	ΔE_{vdW}	ΔG_{SOLV}	ΔG_{nonpol}	ΔG_{bind}
C3	-15.18±0.10	-52.37±0.15	32.84±0.16	-1.25±0.13	**-35.96±0.02**
C4	-12.33±0.10	-63.24±0.05	25.75±0.05	-2.34±0.10	**-52.16±0.20**
C5	-11.96±0.07	-57.84±0.12	36.41±0.02	-3.98±0.30	**-37.37±0.45**
C6	-14.18±0.05	-67.34±0.15	55.81±0.08	-1.55±0.55	**-27.26±0.67**
C7	-11.25±0.10	-73.25±0.02	67.77±0.07	-2.66±0.05	**-19.38±0.10**
Compounds	**NiV-F**				
	ΔE_{ele}	ΔE_{vdW}	ΔG_{SOLV}	ΔG_{nonpol}	ΔG_{bind}
C3	-17.46±0.15	-66.56±0.05	33.51±0.10	-5.36±0.02	**-55.87±0.12**

C4	-10.55±0.05	-43.37±0.25	25.34±0.10	-6.14±0.05	**-34.72±0.25**
C5	-9.86±0.05	-75.55±0.20	56.45±0.22	-7.33±0.10	**-36.29±0.13**
C6	-12.45±0.10	-68.45±0.05	28.66±0.08	-8.33±0.05	**-60.57±0.02**
C7	-10.55±0.20	-43.74±0.02	35.05±0.01	-6.11±0.05	**-25.35±0.26**
Compounds			**NiV-N**		
	ΔE_{ele}	ΔE_{vdW}	ΔG_{SOLV}	ΔG_{nonpol}	ΔG_{bind}
C3	-15.23±0.05	-58.34±0.10	34.23±0.10	-7.36±0.01	**-46.70±0.06**
C4	-12.22±0.02	-61.27±0.07	44.28±0.01	-5.63±0.02	**-17.85±0.10**
C5	-10.28±0.08	-65.84±0.10	38.49±0.05	-1.66±0.02	**-39.32±0.15**
C6	-11.67±0.04	-55.76±0.15	26.34±0.02	-3.66±0.03	**-44.75±0.20**
C7	-13.52±0.03	-62.49±0.12	31.55±0.03	-4.55±0.05	**-49.01±0.17**

Therefore, the study presents five compounds as potential multitarget inhibitors of the Nipah virus, with compound C4 standing out as a particularly strong candidate. The findings suggest the efficacy of computational approaches like molecular docking, dynamics simulations, and binding free energy calculations in the early stages of drug development. These findings contribute to the ongoing efforts to develop effective treatments for Nipah virus, though further experimental validation is necessary before any of these compounds can be considered for clinical use.

Chapter 7: Conclusion & Future Scope

The following are concluding remarks of the study undertaken:

i. The aim of the study was to identify novel potential antiviral hit candidates via application of computational tools such as FBDD, molecular modelling, pharmacokinetics, molecular docking, and molecular dynamics.

ii. The study focuses on the application of FBDD upon FDA-approved antiviral agents/drugs in order to generate novel scaffolds.

iii. The primary objective of the study is to provide comprehensive understanding of the physicochemical characteristics that are essential in the development of drugs. The study demonstrates how necessary it is to apply DFT in order to

comprehend the unique chemical and physical characteristics of the designed hit compounds.

iv. The pharmacokinetic study highlighted the behavior of the designed hit compounds in biological environment. The obtained results were compared with the approved drug and the compounds were screened accordingly.

v. Docking results of the study were found to be comparable, or in some cases better, than the FDA-approved drugs, as they were well explained by a variety of analysis.

vi. The dynamical behavior of the systems was thoroughly examined. By computing various parameters like energy variation, change in radius of gyration, change in number of hydrogen bonds, RMSF, and RMSD, a critical analysis regarding the stability of the drug-receptor complex with evolving time was conducted.

vii. By incorporating the electrostatic effects, the free energy calculations enhanced the computational accuracy of the calculations. Their exhaustiveness also led to more detailed results, such as the energy contributions for each component (van der Waals energy, electrostatic energy, polar solvation energy, and binding energy), as well as the per-residue contribution, which allowed critical analysis of the results.

viii. Overall, the study successfully identified novel potential hit compounds for some common viruses such as Influenza A virus, Variola virus, Ebola virus, and Nipah virus.

The suggestions stipulated for further research:

i. The results obtained in this study can be further investigated against the mutations of different virus species.

ii. We were able to propose a theoretical protocol for carrying out experimental techniques by combining the methods of molecular modelling.

iii. Also, the methods of molecular modelling may be implied to study and develop hit compounds against a wide range of microbial diseases.

Moreover, collaborative molecular modelling studies conducted at the electronic structure level can provide significant insights and invaluable information for the development of new antiviral inhibitors that closely align with experimental research. The ever-improving accuracy of computer-aided drug designing techniques are showing great potential in drug discovery endeavors, and hopefully a panacea for all diseases will be invented in near future.

List of Publications

❖ Papers:

1) **Sinha, P.**, & Yadav, A. K. (2024). Unraveling the anti-breast cancer activity of Cimicifugae rhizoma using biological network pathways and molecular dynamics simulation. *Molecular Diversity*, 1-14.

2) **Sinha, P.**, & Yadav, A. K. (2024). Repurposing integrase inhibitors against human T-lymphotropic virus type-1: a computational approach. *Journal of Biomolecular Structure and Dynamics*, 1-12.

3) **Sinha, P.**, Kumari, R., & Yadav, A. K. (2024). Pd-doped SWCNT as Nanobiosensor for Phenylalanine Hydrolase. In AIP Conference Proceedings, *AIP Publishing*, 3149(1), 030014.

4) **Sinha, P.**, & Yadav, A. K. (2024). Identification of 3, 4-dihydroxy complexes as potential antiviral via DFT, molecular docking, molecular dynamics and MM/PBSA against rabies and dengue receptors. *Journal of Biomolecular Structure and Dynamics*, 42(13), 7037-7053.

5) **Sinha, P.**, & Yadav, A. K. (2024). In silico identification of cyclosporin derivatives as potential inhibitors for RdRp of rotavirus by molecular docking and molecular dynamic studies. *Journal of Biomolecular Structure and Dynamics*, 42(10), 5001-5014.

6) **Sinha, P.**, & Yadav, A. K. (2023). Virtual Screening Based on Docking and Molecular Dynamics Simulations of Potential Ebola Receptor Inhibitors. *ChemistrySelect*, 8(42), e202304059.

7) **Sinha, P.**, & Yadav, A. K. (2023). Molecular docking, molecular dynamics and binding free energy based identification of novel potential multitarget inhibitors of Nipah virus. *Journal of Biomolecular Structure and Dynamics*, 1-17.

8) **Sinha, P.**, & Yadav, A. K. (2023). Computational Analysis on Molecular Stability and Binding Affinity of 3-(Aminothiazolyl) Quinolone Derivative as Multitargeting Antibacterial Agents through Ab Initio Methods and Molecular Docking. *Polycyclic Aromatic Compounds*, 1-22.

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11) **Sinha, P.**, & Yadav, A. K. (2023). Identification of novel potential inhibitor of thymidylate kinase from Variola virus. *Journal of Biomolecular Structure and Dynamics*, 41(23), 14092-14102.

12) **Sinha, P.**, & Yadav, A. K. (2022). Derivatives of 2-methyl tetrahydrofuran-n-carboxylic acids inhibiting novel HA3 subtype of influenza A virus hemagglutinin. *Journal of Computational Biophysics and Chemistry*, 21(07), 783-796.

13) **Sinha, P.**, & Yadav, A. K. (2022). Structural, electronic, spectroscopic and molecular docking analysis of novel hetero oxetane ring compound. *Computational and Theoretical Chemistry*, 1217, 113919.

14) **Sinha, P.**, Pandey, A., & Yadav, A. K. (2020). Vibrational Analysis and Molecular Docking Studies on some Ribonuclease-H HIV Inhibitors. *Biointerface Research in Applied Chemistry*, 11(5), 12796-12807.

❖ Book Chapters:

1) **Sinha, P.**, Kumari, R., & Yadav, A. K. (2024). Unveiling the Transformative Potential of Nanotechnology in Advancing Drug Development. In editors (Singh, S.K., Mishra A.K.) *Applied Research in Physical & Chemical Sciences and Engineering* (pp. 409-432). Lucknow: MKSES Publications & Journals.

2) **Sinha, P.**, & Yadav, A. K. (2024). Antiviral Drug Discovery: Unique Aspects. In editors (Singh, S.K., Shukla, R.K., Gupta, P., Dixit, C.K.) *Interdisciplinary Research in Physical & Chemical Sciences and Engineering* (pp. 136-151). Lucknow: MKSES Publications & Journals.

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4) **Sinha, P.**, & Yadav, A. K. (2023). In-Silico Approaches for Drug Designing and Drug Discovery. In editors (Ansari, H.Q., Singh, S.K., Shukla, R.K., Dixit, C.K.) *Recent Research in Physical & Chemical Sciences and Engineering* (pp. 10-71). Lucknow: MKSES Publications & Journals.

5) **Sinha, P.**, & Yadav, A. K. (2022). Methods of Machine Learning in Cheminformatics and Drug Discovery. In editors (Singh, S.K., Shukla, R.K., Dixit, C.K.) *Advance Research in Physical & Chemical Sciences and Engineering* (pp. 85-110). Lucknow: MKSES Publications & Journals.

❖ Book:

1) A. K. Yadav, **P. Sinha**, R. Kumari (2024) Exploring Quantum Realms: Computational Methods, Synthesis Techniques, and Characterization Tools, *Weser Books*, Edition 1, 978-3-96492-556-5, 134.