# Computer-Aided Drug Design for NS5 Protein of Zika Virus

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## ABSTRACT
Zika virus is an arbovirus that spreads through mosquito bites, sexual transmission, pregnancy and blood transfusions. The infection can be severe leading to birth defects, Guillen- Barre syndrome or it may be mild showing no symptom at all. The present study, aims at using Computer aided drug design to speed up the process of drug discovery by rapid optimization of lead compounds through virtual screening and predicting bioactivity. Docking studies were conducted to screen compounds against NS5 protein of Zika virus. The pharmacokinetic properties of the lead compounds were analysed using online tools.

## General Terms
Computer Aided Drug Design, Virtual Screening, pharmacokinetics

## Keywords
Zika virus, NS5protein, flavonoids,

## 1. INTRODUCTION

### 1.1 NS5 Protein
NS5 or Nonstructural Protein 5 has been found to be an essential component for the replication of Flaviviral Genome [1, 2]. The N terminus of NS5 contains methytransferase. This is connected to RNA dependent RNA polymerase by a short linker. Methytransferase increases RNA synthesis and affects the conformation of polymerase domain. [3, 4, 5] .As it is an essential protein for viral replication it was selected as target protein in the present study [6]

### 1.2 Herbal Compounds
The herbal compounds selected for the study are classified as Plant-based, Mushroom based or known polymerase inhibitors

#### *1.2.1 Plant Based Compounds*
Quercetin is a flavonoid with chemo preventive activity [7]. A research carried out using RNA-containing flaviviruses (Japanese encephalitis, yellow fever, and dengue viruses), bunyaviruses (Punta Toro, sand-fly fever, and Rift Valley fever viruses), alphavirus (Venezuelan equine encephalomyelitis virus), lentivirus (human immunodeficiency virus-type 1) and the DNA-containing vaccinia virus showed that compounds like Lycorine have anti-viral activity. astano spermine and derivatives of Castanospermine have been used to inhibit the Glycosylation of glycoproteins in Rabies virus [8] and it has also been tested against HIV-1 virus [9] .Hesperetin is commonly found flavonoid in citrus fruits such as lemons and oranges. Hesperetin was tested against herpes simplex virus type 1 (HSV-1), polio-virus type 1, parainfluenza virus type 3 (Pf-3), and respiratory syncytial virus (RSV) for its infectivity and replication in the cell. It had no effect on infectivity although it reduced intracellular replication of each of the viruses. [10].The Inhibition of enterovirus 71 replication which was studied using Chrysoplenetin showed that it prevented replication and at the same time it showed low cytotoxicity [11] .Axillarin and Chrysophenol D have been reported to have anti-oxidant and anti-inflammatory actions [12]

#### *1.2.2 Mushroom based compounds*
Ganoderma *pfeifferi* and Ganoderma l*ucidium* extracts have been reported to have antimicrobial activity. Active substances called ganomycins are isolated from the extracts and their derivatives enables their use as antibiotics Epicorazine and Ganodriol are anti-microbial metabolites obtained from Ganoderma species[13] Ganodermatriol has inhibitory actions against HIV-1 protease[14] Lovastatin has shown promising effects against respiratory syntical virus and SARS [15]**.** Hispidin and hispolon showed considerable antiviral activity against influenza viruses' type A and B. Hispidin has also shown inhibitory activity against HIV-1 integrase [16]

#### *1.2.3 Known polymerase inhibitors*
The known polymerase inhibitors used are Cyclophillin Inhibitor which has been Tested against HCV [17] , Acyclovir [18] which is Herpes Simplex Virus DNA Polymerase Inhibitor , Tubercidin [19] and 7-Deaza-2'-C-methyadenosine which possess high antiviral activity and Aphidicolin [20] which is tetracyclic diterpene antibiotic isolated from the fungus *Cephalosporum aphidicola*

## 2. MATERIALS AND METHODS

### 2.1. Protein
Crystal structure of Zika virus NS5 methyl transferase was retrieved from PDB with a ID 5M5B. The crystal structure of the ZIKV MTase was determined at a 2.01-Å resolution in

which one chain is bound to the methyl donor (S-adenosyl-l-methionine [SAM].The SAM binding site **Fig : 1** accommodates a sulfate close to a glycerol that could serve as a basis for structure-based drug design. The crystal structure provides a better understanding of the ZIKV MTase, a central player in viral replication and lay the basis for the development of potential antiviral drugs [21]

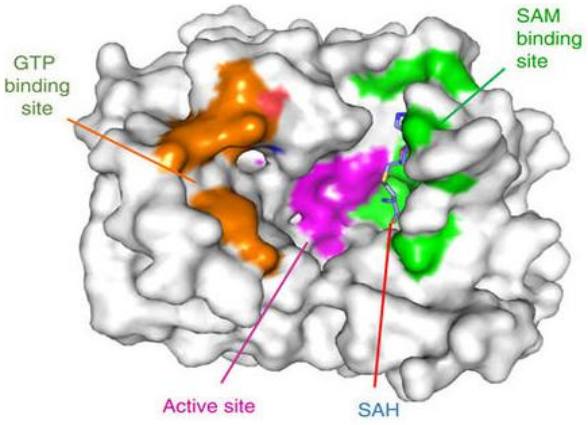


**Fig.1. Structure of NS5 with SAM binding site and GTP binding site (source: Nature)**

## 2.2 Ligands

The structures for the lead compounds were retrieved from NCBI Pub Chem compounds. Compounds with their respective CID's are as follows:

Quercetin-5280343; Lycorine-72378; Hesperetin-72281; Calystegine A3-183073; Calystegine B2-124434; Ganomycin A-9841824; Epicorazine B-73891006; Epicorazine C-10410865; Ganodriol D-15602262; Ganodermatriol-3177; Lovastatin-53232; Hispidin-54685921; Chrysoplenetin-5281608; Chrysoplenol D-5280699; Axillarin-5281603; Cyclophillin Inhibitor-57339992; Tubercidin-6245; Acyclovir -2022; 7-deaza-2'-C-methyladenosine-3011893; Aphidicolin-457964; Celgosivir -60734; Ganoderic Acid A-471002; Lucidumol A-475410; Penduletin-5320462

## 2.3 Active site analysis

S -adenosylmethionine (SAM) was selected to assess the binding site for the present study. The active site residues with hydrogen bonds are identified as Ser 62 and Asp 152 given in Fig.2. Pose view (Fig 3) of PDB was used to perform active site analysis to indicate hydrogen bonds, salt bridges, and metal interactions, hydrophobic interactions and π-π and π-cation interactions. [21]

## 2.4 Physiochemical properties

The physiochemical properties of the retrieved compounds were analysed using Molinspiration property calculator [22]. It was observed that all the compounds selected had no violations according to Lipinski's rule of 5 [23] except Gandodermatriol whose partition coefficient is more than 5 and Ganoderic Acid A with molecular weight greater than 500. The violations have been highlighted in Table.1

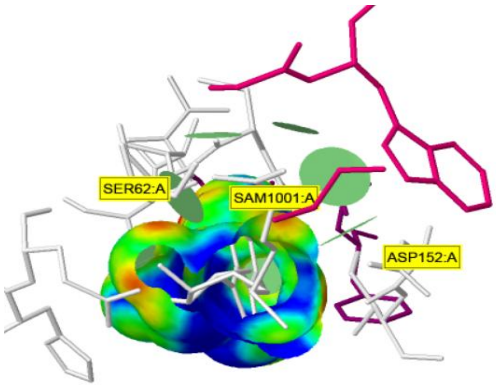


**Fig.2. Active Site residues of SAM in PDB JSMol**

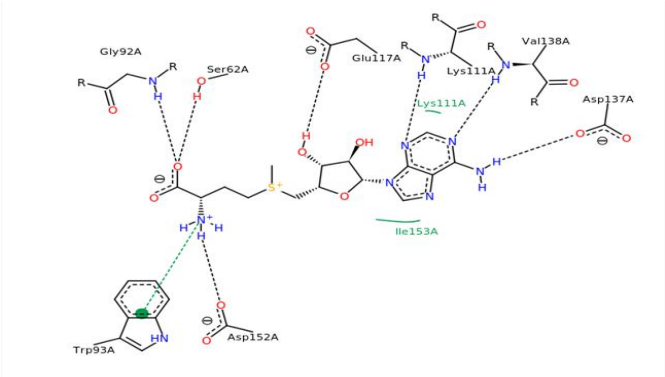


**Fig.3. Pose view Image of SAM in PDB.**

## 2.5 Docking Studies

Protein – Ligand docking was performed using ArgusLab [24, 25].

# 3. RESULTS AND DISCUSSION

## 3.1. Docking Results

The compounds and the binding energy obtained is given in the Table 2 Fig: 4

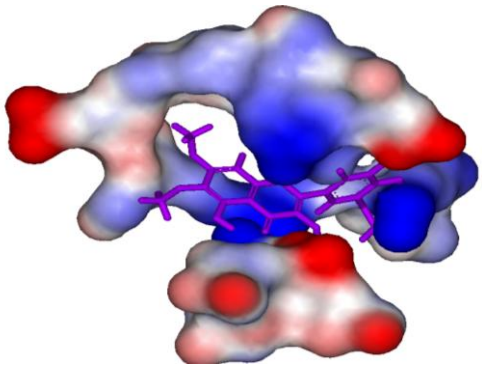

**Fig .4 Best compound Chrysoplenetin in binding pocket**

**Table.1. Molinspiration Results**

| **Name of compound** | **logp** | **TPSA** | **nAtoms** | **MW** | **Hydrogen bond acceptors** | **Hydrogen bond donors** | **rotatable hydrogen bonds** |
|---|---|---|---|---|---|---|---|
| Quercetin | 1.68 | 131.35 | 22 | 302.24 | 7 | 5 | 1 |
| Lycorine | 0.54 | 62.16 | 21 | 287.31 | 5 | 2 | 0 |
| Hesperetin | 1.94 | 96.22 | 22 | 302.2 | 6 | 3 | 2 |
| Castanospermine | -2.04 | 84.15 | 13 | 189.21 | 5 | 4 | 0 |
| Calystegine A3 | -1.26 | 72.71 | 11 | 159.19 | 4 | 4 | 0 |
| Calystegine B2 | -1.96 | 92.94 | 12 | 175.18 | 5 | 5 | 0 |
| Ganomycin A | 4.32 | 97.9 | 26 | 360.45 | 5 | 4 | 10 |
| Epicorazine B | -1.36 | 115.22 | 28 | 420.4 | 8 | 2 | 0 |
| Epicorazine C | -2.25 | 135.4 | 29 | 438.4 | 9 | 3 | 0 |
| Ganoderiol D | 3.94 | 94.9 | 35 | 486.7 | 5 | 3 | 6 |
| Ganodermatriol | 5.1 | 77.75 | 34 | 472.71 | 4 | 3 | 6 |
| Lovastatin | 4.34 | 72.84 | 29 | 404.5 | 5 | 1 | 7 |
| Hispidin | 1.80 | 90.89 | 18 | 246.22 | 5 | 3 | 2 |
| Chrysoplenetin | 2.59 | 107.60 | 27 | 374.31 | 8 | 2 | 5 |
| Chrysoplenol D | 2.28 | 118.60 | 26 | 360.32 | 8 | 3 | 4 |
| Axillarin | 1.98 | 129.89 | 25 | 346.29 | 8 | 4 | 3 |
| Cyclophillin Inhibitor | 1.35 | 86.4 | 23 | 324.4 | 5 | 2 | 7 |
| Tubercidin | -0.93 | 126.66 | 19 | 266.20 | 8 | 5 | 2 |
| Acyclovir | -1.61 | 119.03 | 16 | 225.21 | 8 | 4 | 4 |
| 7-deaza-2'-C-methyladenosine | -1.44 | 126.66 | 20 | 280.28 | 8 | 5 | 2 |
| Aphidicolin | 1.85 | 80.91 | 24 | 338.49 | 4 | 4 | 2 |
| Lucidumol A | 4.95 | 74.60 | 34 | 472.71 | 4 | 2 | 5 |
| Ganoderic Acid A | 2.75 | 128.7 | 37 | 516.67 | 7 | 3 | 6 |
| Penduletin | 2.77 | 98.37 | 25 | 344.32 | 7 | 2 | 4 |
| Celgosivir | -0.41 | 90.23 | 18 | 259.3 | 6 | 3 | 4 |

**Table.2. Docking results**

| Name Of The Compound | Binding Energy (Kcal/mol) |
|---|---|
| Gandoderic A | -5.56352 |
| Lovastatin | -5.7225 |
| Acyclovir | -5.91259 |
| Tubercidin | -6.74719 |
| Cyclophillin | -7.20181 |
| Lycorine | -7.27243 |
| Epicorazine C | -7.2865 |
| Epicorazine B | -7.34809 |
| 7'Deazamethlyadenosine | -7.36634 |
| Celgovisir | -7.39762 |
| Calystegine B2 | -7.43899 |
| Chrysoplenetin | -7.57758 |
| Pendulitin | -7.73345 |
| Hesperetin | -7.84153 |
| Luidumol A | -7.89849 |
| Castanospermine | -7.92129 |
| Quercetin | -8.09155 |
| Axillarin | -8.10503 |
| Hispidin | -8.40889 |
| Ganodermanontrol | -8.51077 |
| Calystegine A3 | -8.54332 |
| Ganoderil D | -8.58893 |
| Cyclophillin | -8.70669 |

## 3.2 Pharmacokinetics

Absorption, Distribution, Metabolism, Excretion and Toxicity (ADMET) properties of the compounds were analysed using pkcsm [26, 27] .The properties that were looked into are Blood Brain permeability, CNS permeability, substrate for cytochrome, and substrate for Renal OCT2, Absorption in Human Intestine, and Hepatotoxicity None of the first ten compounds cross the Blood brain barrier or CNS.. Lovastatin, Cyclophillin, Lycorine and Chrysoplenetin were found to be substrates for cytochrome. Lovastatin was the only compound that is a substrate for Renal Organic Chain Transporter (Renal OCT2).The compound that could possibly be poorly absorbed by the Human Intestine are Calystegine B2 (44%), followed by Celgovisir (47%), and acyclovir (48%). The lead compounds that could be highly absorbed are Chrysoplenetin (99.8%), Cyclophillin (96%), and lovastatin (94%)

# 4. CONCLUSION

Docking studies were conducted to identify the lead compounds which were effective against NS5 of Zika virus. Based on binding energies and ADMET studies it can be concluded that Lovastatin, Acyclovir, Chrysoplenetin, Epicorazine B and C can be considered as suitable drugs to cure Zika virus infection. Among these the best compound identified based on binding affinity and active site analysis is Chrysoplenetin.

# 5. ACKNOWLEDGMENTS

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