BIOINFORMATION
Discovery at the interface of physical and biological sciences

open access www.bioinformation.net Hypothesis

Volume 11(7)

# Molecular docking based screening of neem-derived compounds with the NS1 protein of Influenza virus

**Aftab Ahmad*, Ammara Ahad, Abdul Qayyum Rao & Tayyab Husnain**

Center of Excellence in Molecular Biology (CEMB), University of the Punjab, West Canal Road, 53700, Lahore, Pakistan; Aftab Ahmad - Email: warraich6229@cemb.edu.pk; Phone: +92-(0)302-6416094; *Corresponding author

Received May 18, 2015; Revised June 09, 2015; Accepted June 10, 2015; Published July 31, 2015

**Abstract:**
Different strains of influenza virus are affecting a large number of people worldwide to combat with Influenza virus destruction, numerous synthetic antiviral medicines are available for influenza virus in the market. But still there was a need for the development of drug which will target all the strains of influenza virus. For this purpose conserved residues within the influenza virus NS1 protein have been found by aligning all the available sequences of existing strains from the national center of biotechnology information(NCBI) protein database. The compounds from leaf extracts of neem (*Azadirachta indica*), previously known to have antiviral properties, were virtually screened to identify side effects free natural drug. Molecular docking identified eight potential compounds (Tetratriacontane, 127-40-2, 6-o-ACETYLNIMBANDIOL, Rutin, Tiplasinin, Hyperoside, ( )-Nimocinolide and Quercitrin) found to have perfect binding with reported conserved residues (R19, R35, S42 and D39) of influenza virus NS1 protein involved in the binding of drugs. From, further analysis 6-o-ACETYLNIMBANDIOL, Rutin and Tiplasinin were found as drug against influenza strains because their binding residues were conserved in all strains. The potential of neem chemical against influenza virus has best been highlighted through this study and it provides direction for further consideration of these products for in-vivo and in-vitro validations.

**Key Words**: Influenza virus; NS1 protein; Neem leaf extract; Molecular docking;

**Abbreviations:** NS1 protein; Non Structural 1 protein; NA Neuraminidase; HA Hemagglutinin; M Matrix; 127-40-2 4-[(1E, 3E, 5E, 7Z, 9E, 11E, 13E, 15E, 17E)-18-(4-hydroxy-2,6,6-trimethylcyclohex-2-en-1-yl)-3,7,12,16-tetramethyloctadeca-1,3,5,7,9,11,13,15,17-nonaenyl]-3, 5, 5-trimethylcyclohex-3-en-1-ol; Quercitrin2- (3,4-dihydroxyphenyl)-5,7-dihydroxy-3- [(2S,3R,4R,5R,6S)-3,4,5-trihydroxy-6-methyloxan-2-yl]oxychromen-4-one; Tiplasinin 2-[1-benzyl-5-[4-(trifluoromethoxy) phenyl] indol-3-yl]-2-oxoacetic acid; Hyperoside2-(3,4-dihydroxyphenyl)-5,7-dihydroxy-3- [(2S,3R,4S,5R,6R)-3, 4, 5-trihydroxy-6- (hydroxymethyl)oxan-2-yl]oxychromen-4-one LGH4-(2-chloro-4-nitrophenyl)piperazin-1-yl][3-(2-methoxyphenyl)-5-methyl-1,2-oxazol-4-yl]methanone; nRUTIN2-(3, 4-dihydroxyphenyl) -5, 7-dihydroxy-3-[(2S, 3R, 4S, 5S, 6R)-3, 4, 5-trihydroxy-6-[[(2R, 3R, 4R, 5R, 6S)-3,4,5-trihydroxy-6-methyloxan-2-yl]oxymethyl]oxan-2-yl]oxychromen-4-one.

**Background:**
Influenza A viruses are common pathogens having high variability that caused acute respiratory disease and resulted in high death rates (20 to 40 million people) worldwide till 1918 **[1].** The influenza A viruses (H5N1) are violently spreaded in Southeast Asia in present years so they are robust candidates for causing the flu pandemic **[2].** Influenza is a significant health problem due to its rapid transmission and high mortality rate. Influenza is a respiratory infection caused by the influenza virus **[3]** belonging to the family Orthomyxoviridae. The virus has single stranded and segmented RNA genome which encodes for 8 proteins. For the

 

propagation of influenza virus the NS1 protein, matrix (M1, M2), neuraminidase (NA), hemagglutinin (HA) and three viral polymerase subunits (PB1, PB2 and PA) plays an active role **[4].** Influenza virus is classified into three groups and has more than 10 strains. Its genome is highly variable with many strains having a high rate of mutation which make them resistant to many drugs. Previously the disease was cured by either using nucleic acid protein inhibitors (e.g. Zanamivir), neuraminidase inhibitors (e.g. Zanamivir, Oseltamivir), ion channel blockers (e.g. Amantadine, Rimantadine) or siRNA technique **[5]**. The whole concept of the gene regulation and proteome function can be used to predict antiviral drug target of projected motifs of influenza A virus **[6]**. Host specific epitope of influenza A virus surface proteins NA and HA, has been predicted which could support in designing the drugs against Influenza virus **[7].** Oseltamivir and Zanamivir have been proved as an efficient inhibitors of NA. Due to the mutation in the active site of NA and HA, resistance of influenza virus against these drugs has been reported **[8]**.

Reasons for H5N1 subtype influenza A virus virulence is unclear yet, but some causative aspects have been recognized. The non-structural (NS) protein 1 is one of them **[9]**. The transcription and replication of influenza virus RNAs took place in the nucleus of the infected cells **[10].** Genome of influenza A viruses encoded ten to eleven viral proteins from eight single-stranded negative sense RNA segments depending on the strain. Except for NS1 and PB1F2, all others were structural proteins. The NS1 and NS2 proteins were translated from differential splicing mRNA of segment 8, whereas NS2 was nuclear export protein. The RNA segment 8 of influenza A virus, had 890 nucleotides, which directed the synthesis of 2 mRNAs in infected cells. The NS1 protein consisted of 230 amino acids; while the other derived from alternative splicing of NS1 mRNA was NS2 protein having 121 amino acids **[11]**. This RNA binding protein was involved in controlling post transcriptional processing steps i.e Inhibition of nuclear pass of mRNAs that contain 3′ poly (A) and pre-mRNA splicing **[12].**

Medicinal plants are very important for treatment of different diseases, mainly in the countries where there are insufficient resources. Use of traditional medicines are mainly encouraged in most of the world population **[13].** These traditional medicines have fewer side effects than other allopathic medicines, one of the major reasons to isolate and process these compounds from plants **[14].** *Azadirachta indica* (neem) is a medicinal plant and has grown universal importance in recent years. Neem has been extensively used in Ayurveda, Unani, and Homeopathic medicines. It has a huge range of biologically active chemicals that are chemically and structurally different. From different parts of this plant i.e. flowers, leaves, seeds, roots, fruits and bark more than 140 chemically active compounds have been isolated and are being used traditionally as a cure for many diseases. These active compounds have been identified as an anti-inflammatory, anti-ulcer, anti-hyperglycaemic, immune-modulatory, anti-mutagenic, anti-carcinogenic, anti-oxidant, and anti-viral drugs **[15]**.

Neem elements are mainly divided in two groups: Non-isoprenoids and Isoprenoids. The non-isoprenoids comprise of proteins, sulphurous compounds, carbohydrates and polyphenolics including dihydrochalcone, flavonoids, coumarin, and aliphatic compounds. The isoprenoids consist of di-terpenoids and tri-terpenoids which include azadirone, protomeliacins, limonoids and some derivatives such as nimbin, vilasinin, salanin and azadirachtin **[13].** By an alcoholic extract of neem leaves a dose dependent substantial decrease in blood pressure has also been reported **[16]**.

In the light of above debated medications by neem we designed this study to screen *Azadirachta indica* (Neem) active compounds against the influenza virus NS1 protein through molecular docking to study their interaction pattern. It was also observed that either these compounds were interacting with conserved residues are not. From this study it was concluded that either these compounds could be act as competent drug.

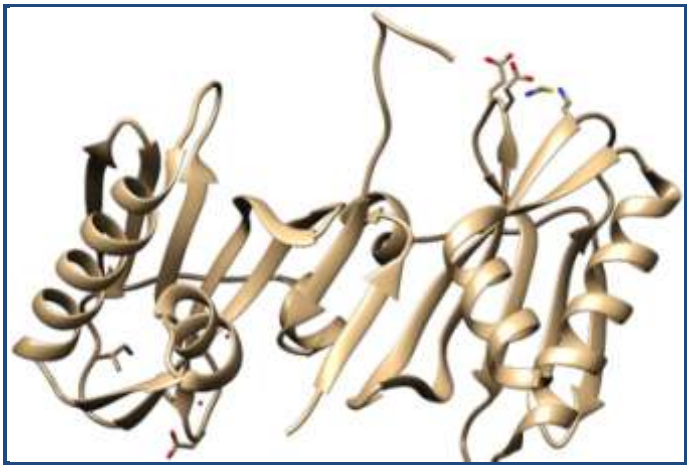

**Figure 1:** NS1 effector domain structure and legends attached to the model, taken from PDB with PDB ID (2GX9)

**Methodology:**

*In-silico* analysis of active chemicals of *Azadirachta indica* (neem) leaves against influenza virus NS1 protein was carried out. For this investigation, chemical structures of compounds of neem leaves were retrieved in a MOL format from chemical database PubChem available on NCBI website. Some of the chemical compounds were drawn in MOL format in Chemdraw software. NS1 protein structure (PDB ID: 2GX9) used for docking purpose was downloaded from Protein Data Bank (PDB) as shown in **Figure 1.** For molecular docking analysis, Molecular Operating Environment (MOE) software was used **[17].**

***Finding the NS1 protein conserved residues within and among the influenza strains:***

In order to find the conserved residues of NS1 protein within and among the strains of influenza virus active against humans (i.e. H1N1, H1N2, H2N2, H3N2, H5N1, H7N2, H7N3, H7N7, and H9N2), the available NS1 protein sequences of each strain were retrieved from NCBI protein database. At first, the retrieved NS1 protein sequences of each strain were aligned by multiple alignment through Clustal Omega (http:// www.ebi.ac.uk/ Tools/msa/clustalo/) to develop a consensus sequence for each strain, then by using the CLC Genomics Workbench 8 conserved residues within the strain were identified. The conserved residue consensus sequences of all

the strains were again aligned using the CLC Genomics Workbench 8 to get the final conserved consensus sequence among the above said strains **(Figure 2).**

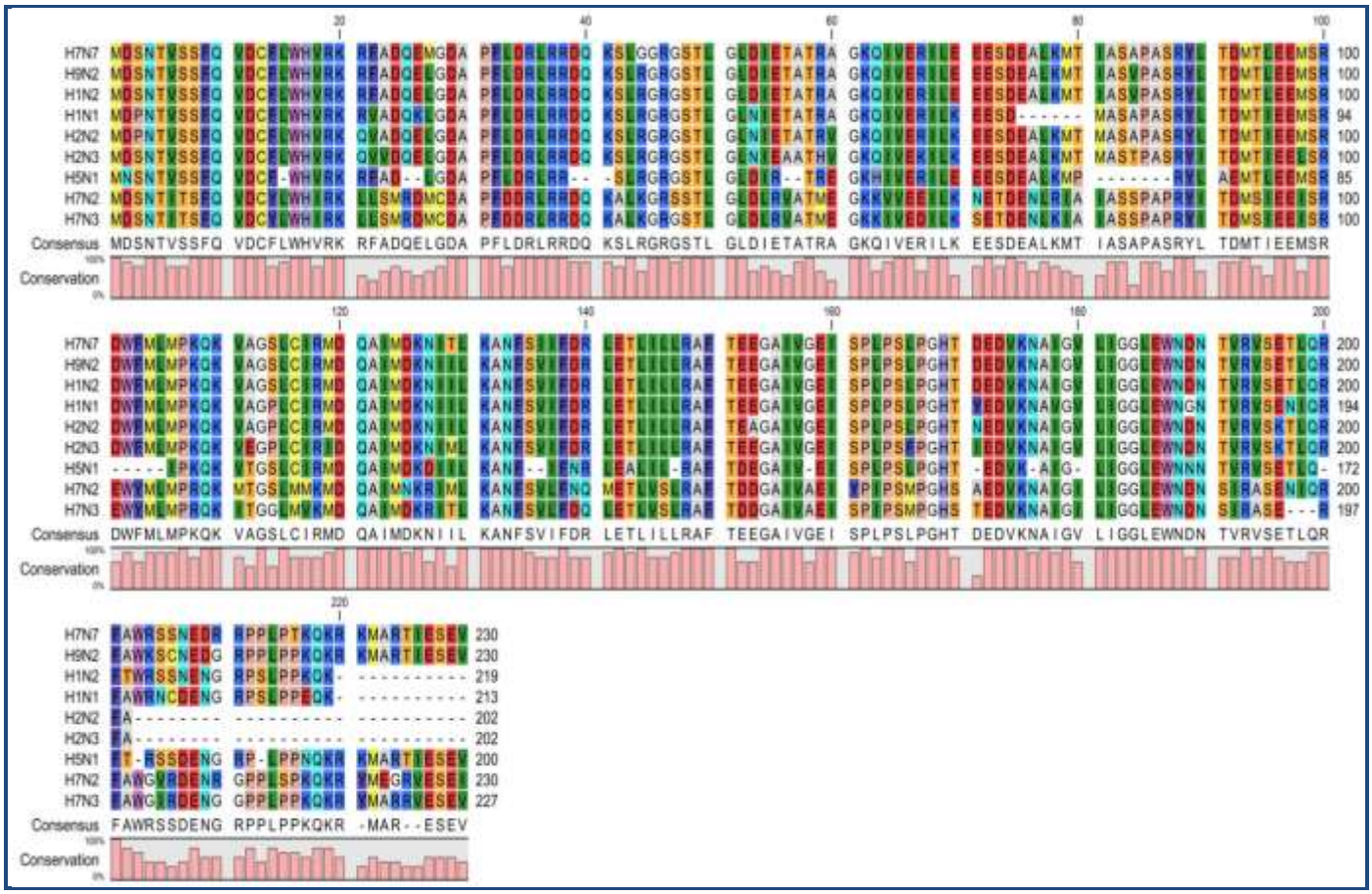


**Figure 2:** Multiple sequence alignment of influenza virus NS1 protein consensus sequences of each strain (i.e. H5N1, H7N2, H7N3, H9N2, H7N7, H1N1, H2N3, H1N2 and H2N2) using CLC Genomics Workbench 8. For the development of each consensus sequence, all the available NS1 protein sequences of the above said strains were retrieved from NCBI database and were converted to consensus sequences using CLC Genomics Workbench 8. The colored bars at the bottom are representing the conservation level. Conservation was 43 %.

**Molecular Docking:**

***Preparation of Protein Structure (Receptor)***

Three-dimensional model of target protein of influenza virus was retrieved from PDB [PDB ID: 2GX9, 2GX9 is a full-length NS1 effector domain 3D model available in PDB. All the water molecules were removed and hydrogen atoms were added by using MOE software. Optimization of receptor molecule was done by energy minimization and 3D protonation using AMBER99 force field option of MOE. The gradient was 0.05 and receptor was minimized unless root mean square gradient reached below 0.05. After that the receptor protein was 3D protonated and then hydrogen molecules were hidden by using hide molecule option. dsRNA binding site 5-50 residues of NS1 protein were selected as a pocket site to destroy the dsRNA binding activity of the NS1 protein. Surface and maps option of MOE was used to point out the surface of the docking site and pocket residues. This energy minimized and 3D protonated receptor molecules were then used for docking analysis.

***Preparation of ligand structure and construction of database***

The structures of biologically active compounds of neem leaves were downloaded from the PubChem database in 2D format **Table 1 (see supplementary material).** Some structures of chemical compounds were not presented in the PubChem database so their 2D structures were retrieved from literature study and were drawn in 3D format by using ChemDraw software. For preparation of ligand structures for docking, hydrogen atoms were added to each ligand and their energy was minimized by using the MMFF94X force field at 0.05 gradients. Then these ligand structures were saved in .mol2 file format. The database was created and saved in .mdb format which was used for docking against the target receptor protein.

***Docking analyses***

After preparation of receptor protein and ligand molecules, molecular docking was executed against the databases mentioned earlier. Docking output database file which contains receptor ligand complex was saved in .mdb format. The docked complexes were sorted with respect to increasing S value (the final score to indicate binding free energy). The complexes with minimum S were taken to evaluate the interactions of ligand with the active site residues of the receptor protein. The best hydrogen bonding and π-π interactions were analyzed by the ligX option of MOE.

**Results & Discussion:**

***Finding conserved residues within and among influenza virus NS1 protein***

To find the conserved residues in all the strains of influenza virus **(Figure 2),** alignment was done by using the CLC Genomics Workbench 8 as described earlier. In the **Figure 2** conservation of residues is shown by vertical bars. The alignment showed that NS1 protein is a moderate conserved protein (43.47%).

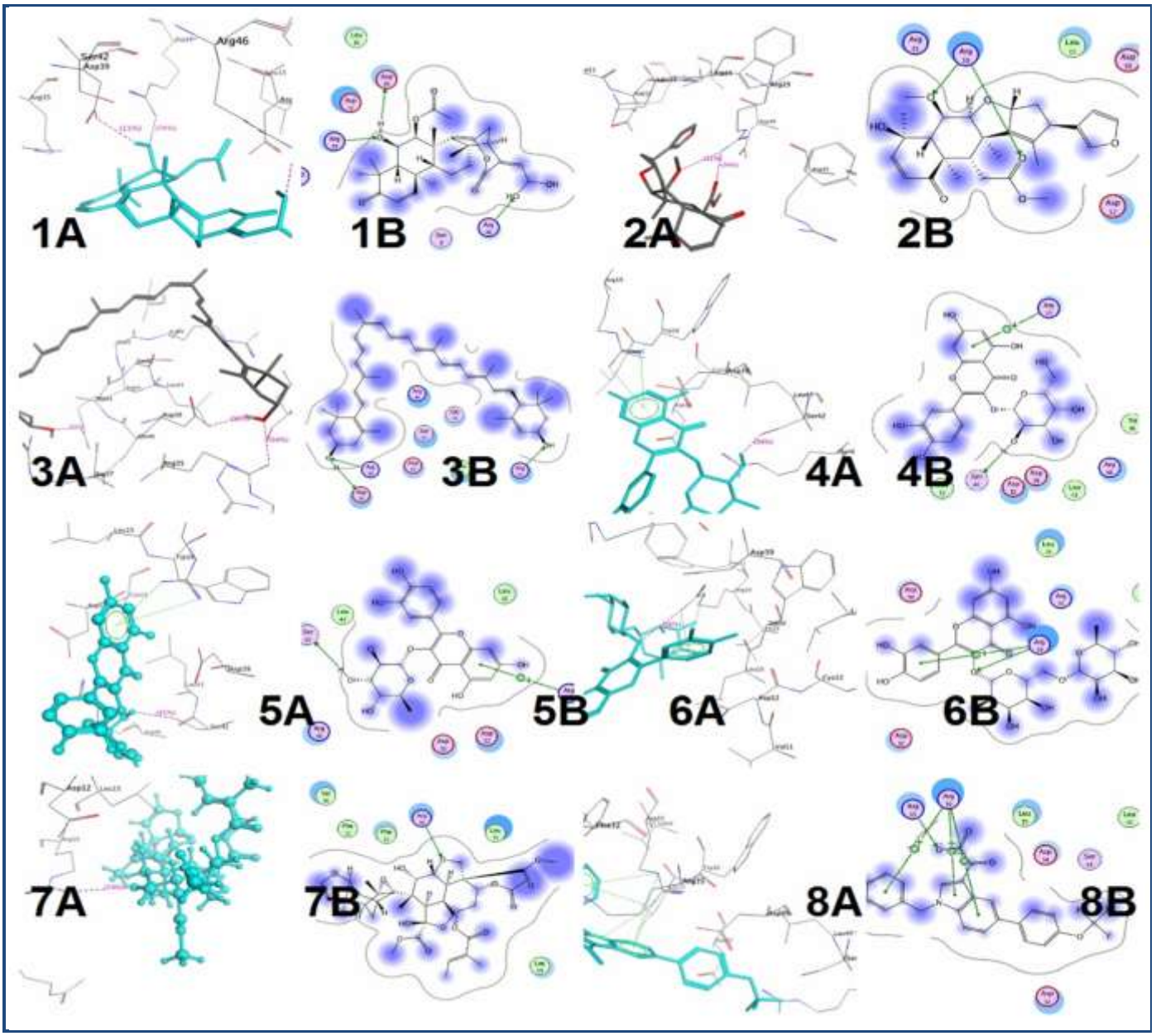


**Figure 3***:* Interaction diagrams of phytochemicals from neem leaf extract compound with influenza virus NS1 protein. Where; 1A & 1B are two dimensional and three dimensional interaction diagrams of R21, V18, W16 and C13 residues of influenza virus NS1 protein with Tetratriacontane, respectively; 2A & 2B are showing the interaction of R19, R44, and D39 residues of NS1 protein with 127-40-2, 3A and 3B part of the diagram is illustrating the binding between R19 residue with 6-o-ACETYLNIMBANDIOL. Furthermore, 4A and 4B is illuminating quercitrin and R19, S42 residue interactions, 5A and 5B shows Tiplasinin, 6A and 6B shows Hyperoside, 7A and 7B showing RUTIN, 8A and 8B is showing ( )-Nimocinolide interaction with NS1 protein residues. Interaction diagrams were attained by using ligand interaction analysis feature of MOE

***Docking analyses against Azadirachta indica (neem) leaf chemicals***

Docking of influenza virus NS1 protein against *Azadirachta indica* (neem) leaf chemicals resulted in 8 complex conformations. Tetratriacontane showed least S-score and interactions with the R21, V18, W16, and C13 residues of NS1 protein **Table 2 (see supplementary material); Figure 3(1A & 1B).** The other eight compounds were also having a lower S-score and strong hydrogen bonds with multiple residues of the NS1 protein **Figure 3 (2A & 3B).** All these compounds showed interactions with R19 residue of selected pocket of the NS1 protein. R44 showed interaction only with 4-

[(1E,3E,5E,7Z,9E,11E,13E,15E,17E)-18-(4-hydroxy-2,6,6-trimethylcyclohex-2-en-1-yl)-3,7,12,16-tetramethyloctadeca-1,3,5,7,9,11,13,15,17-nonaenyl]-3,5,5-trimethylcyclohex-3-en-1-ol(127-40-2) while D39 showed the interactions with (127-40-2) and ( )-Nimocinolide. S42 interacted with 2-(3,4-dihydroxyphenyl)-5,7-dihydroxy-3-[(2S,3R,4R,5R,6S)-3,4,5-trihydroxy-6-methyloxan-2-yl]oxychromen-4-one(Quercitrin) and 2-(3,4-dihydroxyphenyl)-5,7-dihydroxy-3-[(2S,3R,4S,5R,6R)-3,4,5-trihydroxy-6-(hydroxymethyl)oxan-2-yl]oxychromen-4-one(Hyperoside). R35 was found in complex with 2-[1-benzyl-5-[4-(trifluoromethoxy) phenyl] indol-3-yl]-2-oxoacetic(Tiplasinin) and R46 with ( )-Nimocinolide. 6-o-ACETYLNIMBANDIOL Hyperoside and Quercitrin. 6-o-ACETYLNIMBANDIOL and Rutin were having only one interaction with R19.

***Conserved residues of NS1 protein involved in interaction with Azadirachta indica (neem) leaf chemicals***

The docking showed that R21, V18, W16, C13, R19, S42, R44, D39, R35 and R46 were residues involved in the interaction with the neem leaf chemicals. W16, C13, R19, R35 and R46 were found to be conserved residues in the NS1 protein of Influenza virus. D39 was deleted in H5N1 influenza strain. From Table-2, it is obvious that 6-o-ACETYLNIMBANDIOL, Rutin and Tiplasinin have shown strong interactions with only R19 and R35, which both are conserved residues, so these drugs can be used against influenza virus. ( )-Nimocinolide was having interactions with R19, R46, D39. D39 was absent in H5N1 influenza strain so, on the basis of these results its use as antiviral drug against all the influenza virus strains except H5N1 has been suggested. S43 was mutated with A42 in both H7N2 and H7N3. So Quercitrin and Hyperoside can be used as a drug except H7N2 and H7N3. S42 and A42 have similar group and similar function so it is hypothesized that there is a chance that both drugs can be used against H7N2 and H7N3 influenza strains.

**Discussions:**

The alignment showed that NS1 protein is a moderate conserved protein (43.47%). NS1 is not a conserved protein, but it has many residues which are conserved in all the Influenza strains. Docking analysis showed that interacting compounds from neem mostly interacted with R19, which is a conserved residue. R21, V18, W16, C13, R19, S42, R44, D39, R35 and R46 these residues of the NS1 protein were involved in interaction with neem compounds. Tetratriacontane, 127-40-2, Tiplasinin, Hyperoside, RUTIN, ( )-Nimocinolide, 6-o-ACETYLNIMBANDIOL and Quercitrin were neem compounds which gave best interacrions with NS1 of influenza virus. From these compounds 6-o-ACETYLNIMBANDIOL, Rutin and Tiplasinin were involved in interaction only with conserved residues so they can be used as drug against all the influenza strains.

One of interacting residue D39 of ( )-Nimocinolide was absent in H5N1so, it cant be used against that strain. One of interacting residue SER43 of Quercitrin and Hyperoside was mutated with A42 in both H7N2 and H7N3 so, these both drugs cannot be used against these strains.

All these interacting compounds were found to have antiviral activities and were reported as Rutin has been reported previously to have strong antiviral, antimicrobial and antifungal activity **[18]**. Hyperoside also has antiviral activity which is also previously reported against hepatitis B virus (HBV) **[19]**. Nimbaflavone is also tested for antiviral activity **[20].** Thr5, Pro31, Asp34, R35, Arg38, Lys41, Gly45, R46 and Thr49 are reported resides of NS1 protein involved in the attachment of the dsRNA. R46 and R35 same residues, D39 and R44 neighboring residues involved in the interaction with our reported drugs. So by using these drugs dsRNA binding activity of the NS1 protein can be blocked **[21, 22]**.

Due to this blockage NS1 protein will be unable to block the activation of protrin kinase. phosphorylates the alpha subunit of eukaryotic translation initiation factor 2 (elF-2 alpha), leading to a decrease in the rate of initiation of translation. In the absence of NS1, this pathway is inhibited during anti-viral response to halt all protein translation – thus stopping the synthesis of viral proteins; however, the influenza virus' NS1 protein is an agent that circumvents host defenses to allow viral gene transcription to occur.

**Conclusion:**

Due to strain variations among influenza virus, it is the need of the time to identify the conserved residues among different strains as a target for drug discovery based on compounds extracted from natural sources like the *Azadirachta indica* (neem) leaves. For this study NS1 protein was selected and screened against compounds extracted from neem leaves using *in-silico* screening and molecular docking simulation techniques. The compounds 6-o-ACETYLNIMBANDIOL, Rutin, Tiplasinin, Hyperoside, ( )-Nimocinolide and Quercitrin showed best interactions with conserved residues of the NS1 protein. So these compounds have been identified for holding great potential for utilization as a drug against influenza strains. These observations require further considerations for *in-vivo* and *in-vitro* validations. The above reported drugs are screened from natural source and are having less or no side effect as compared to synthetic drug compounds. These screened drugs can be further synthesized and validate in wet lab against all the strains of Influenza virus.

**Acknowledgement:**

The Higher Education Commission (HEC) is highly acknowledged for providing the financial support to carry out the proposed research. The authors are also grateful for the contributions of members of the Protein Engineering Section, Bioprocess Engineering Lab, Department of Bioinformatics and Biotechnology, Government College, University Faisalabad (GCUF) and Plant Biotechnology laboratory, Centre of Excellence in Molecular Biology (CEMB), University of the Punjab Lahore, Pakistan.

**References:**

**[1]** Taubenberger JK & Morens DM, *Emerg Infect Dis*. 2006 **12**: 15 [PMID: 16494711]
**[2]** Horimoto T & Kawaoka Y, *Trends Mol Med*. 2006 **12**: 506 [PMID: 17011235]
**[3]** Body M *et al. Med J Aust*. 2006 **20**: 185 [PMID: 17115951]
**[4]** Kamal RP *et al. Arch Virol*. 2007 **152**: 1637 [PMID: 17558541]
**[5]** Stiver G, *CMAJ*, 2003 **168**: 49 [PMID: 12515786]
**[6]** Somvanshi P *et al. OMICS*. 2008 **12**: 61 [PMID: 18266559]

 

**[7]** Somvanshi P *et al. OMICS*. 2008 **12**: 61 [PMID: 18266559]
**[8]** Nicholson KG *et al. Lancet*. 2003 **362**: 1733 [PMID: 14643124]
**[9]** Solórzano A *et al. J Virol*. 2005 **79**: 7543 [PMID: 15919908]
**[10]** Fortes P *et al. The EMBO J*. 1994 **13**: 704 [PMID: 8313914]
**[11]** Bullido R *et al. J Virol*. 2001 **75**: 4912 [PMID: 11312364]
**[12]** Nemeroff ME *et al. Mol Cell*. 1998 **1:** 991 [PMID: 9651582]
**[13]** Jalil A *et al. Bioinformation* 2013 **9:** 1031 [PMID: 24497731]
**[14]** Akanksha K *et al. Indian J Exp Biol*. 2010 **48**: 294 [PMID: 21046984]
**[15]** Subapriya R & Nagini S, *Curr Med Chem Antic ancer Agents*. 2005 **5:** 149 [PMID: 15777222]
**[16]** Koley KM & Lal J, *Indian J Physiol Pharmacol*. 1994 **38**: 225 [PMID: 7814089]
**[17]** Khan M *et al. Bioinformation* 2013 **9**: 710 [PMID: 23976825]
**[18]** Orhan DD *et al. Microbiol Res*. 2010 **165**: 496 [PMID: 19840899]
**[19]** Wu LL *et al. Acta Pharmacol Sin*. 2007 **28**: 404 [PMID: 17303004]
**[20]** Gangar SC *et al. Acta Pol Pharm*. 2010 **67:** 381 [PMID: 20635534]
**[21]** Wang W *et al. Rna*. 1999 **5:** 195 [PMID: 10024172]
**[22]** Yin C *et al. J Biol Chem*. 2007 **282**: 20584 [PMID: 17475623]

**Edited by P Kangueane**
**Citation: Ahmad *et al.*** Bioinformation 11(7): 359-365 (2015)

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## Supplementary material:

**Table 1:** Drugs common names and PubChem ID along with drugs IUPAC name or chemical name

| No | Drugs Common name **and PubChem ID** | Drugs IUPAC name or Chemical name |
|---|---|---|
| 1 | quercitrin(5280459) | 2-(3,4-dihydroxyphenyl)-5,7-dihydroxy-3-[(2S,3R,4R,5R,6S)-3,4,5-trihydroxy-6-methyloxan-2-yl]oxychromen-4-one (5280459) |
| 2 | Tiplasinin(6450819) | 2-[1-benzyl-5-[4-(trifluoromethoxy)phenyl]indol-3-yl]-2-oxoacetic acid |
| 3 | Hyperoside(5281643) | 2-(3,4-dihydroxyphenyl)-5,7-dihydroxy-3-[(2S,3R,4S,5R,6R)-3,4,5-trihydroxy-6-(hydroxymethyl)oxan-2-yl]oxychromen-4-one |
| 4 | RUTIN(5280805) | 2-(3,4-dihydroxyphenyl)-5,7-dihydroxy-3-[(2S,3R,4S,5S,6R)-3,4,5-trihydroxy-6-[[(2R,3R,4R,5R,6S)-3,4,5-trihydroxy-6-methyloxan-2-yl]oxymethyl]oxan-2-yl]oxychromen-4-one |
| 5 | Tetratriacontane(26519) | Tetratriacontane |
| 6 | ( )-Nimocinolide(6442906) | Nimocinolide; ( )-Nimocinolide; 104494-22-6; 24-Norchola-1,14,20(22)-trien-21-oic acid, 7-(acetyloxy)-6,23,23-trihydroxy-4,4,8-trimethyl-3-oxo-, gamma-lactone, (5alpha,6alpha,7alpha,13alpha,17alpha)-; |
| 7 | 127-40-2(46835684) | 4-[(1E,3E,5E,7Z,9E,11E,13E,15E,17E)-18-(4-hydroxy-2,6,6-trimethylcyclohex-2-en-1-yl)-3,7,12,16-tetramethyloctadeca-1,3,5,7,9,11,13,15,17-nonaenyl]-3,5,5-trimethylcyclohex-3-en-1-ol |
| 8 | 6-O-acetylnimbandiol | 6-O-acetylnimbandiol; CHEBI:67306; methyl [(2R,3aR,4aS,5R,5aS,6R,9aR,10S,10aR)-5-(acetyloxy)-2-(furan-3-yl)-6-hydroxy-1,6,9a,10a-tetramethyl-9-oxo-3,3a,4a,5,5a,6,9,9a,10,10a-decahydro-2H-cyclopenta[b]naphtho[2,3-d]furan-10-yl]acetate; |

**Table 2:** Docking score (S) and interaction sites of neem phytochemicals against influenza virus NS1 protein.

| No | Drug IUPAC Name and Pubchem ID | Score (S) | Interacting Residues |
|---|---|---|---|
| 1 | Tetratriacontane | -19.3730 | ARG21, VAL18, TRP16, CYS13 |
| 2 | 127-40-2 | -19.3377 | ARG19, ARG44, ASP39 |
| 3 | 6-o-ACETYLNIMBANDIOL | -18.6482 | ARG19 |
| 4 | Quercitrin | -17.8655 | ARG19, SER42 |
| 5 | Tiplasinin | -17.1605 | ARG19, ARG35 |
| 6 | Hyperoside | -16.3806 | ARG19, SER42 |
| 7 | RUTIN | -16.7127 | ARG19 |
| 8 | ( )-Nimocinolide | -15.0279 | ARG19, ARG46, ASP39 |