International Journal of Science and Research (IJSR)
ISSN (Online): 2319-7064
Index Copernicus Value (2016): 79.57 | Impact Factor (2015): 6.391

Docking and Inhibition of Antiviral Ligands of Japanese Encephalitis Virus with Capsid Envelope Strain SA-14-14-2(3P54)

Managobinda Rath[1], Dr. Mahesh Chanrda Sahu[1], Dr. Susant Kumar Sahu[2]

[1]Directorate of Medical Research, IMS & SUM Hospital, Siksha O Anusandhan University, Bhubaneswar, K8, Kalinga Nagar, Bhubaneswar-751003.

[2]Department of Pharmacy, Utkal University, Bhubaneswar, Vani Vihar, Bhubaneswar-751004

Abstract: ***Japanese Encephalitis Virus (JEV) is a flavivirus that threatens more than half of the world's population. This disease is most prevalent in Southeast Asia and East Asia and mortality is generally much higher in children. Lifelong neurological defects such as deafness, emotional liability and hemiparesis may occur in those who have had central nervous system involvement. Genomic arrangement of the virus shows a single-stranded RNA genome packaged in the capsid which is formed by the capsid protein. The outer envelope is formed by envelope protein and is the protective antigen. It aids in entry of the virus into the cell. The genome also encodes several nonstructural proteins (NS1, NS2a, NS2b, NS3, N4a, NS4b, and NS5). There is no specific antiviral drug is yet available for clinical therapy. The tertiary structure of protein 3P54 was found in Protein data bank(PDB) and the structure was refined by Pymol and inhibitors were found from different literature study. Three Inhibitors of Capsid Envelope protein have been screened by applying Lipinski's Rule of five then docked against Capsid Envelope protein of JEV. Different Dope score have been recorded for different ligands. However cilnidipine, niclosamide forming hydrogen bond and FGIN-1-27 is not forming hydrogen bond atom during the docking process against the target protein 3P54. Further study on this aspect may help to discover more effective drugs for the treatment of this deadly disease caused by Japanese Encephalitis Virus.***

Keywords: Flavivirus, JEV Envelope Protein, Ligands, Lipinski's Rule of five, Docking, pymol, discovery Sudio4.0

1. Introduction

Japanese Encephalitis is now one of the most concern disease in Asia, especially in rural and suburban areas where rice culture and pig farming coexist. Japanese Encephalitis (JE) is a mosquito-borne flavivirus viral disease and also known as Japanese B encephalitis to distinguish from Economo's A encephalitis. Japanese encephalitis including West Nill Virus (WNV), Tick-Borne Encephalitis Virus (TBEV) and Dengue Virus (DV) transmission cycle is Culex tritaeniorhynchus mosquitoes and similar species that lay eggs in rice paddies and other water sources, with pigs, wild birds and aquatic birds but may also be transmitted to humans and horses. It is affecting 30, 000 to 50, 000 cases and 10, 000 deaths in eastern Asia per year. There are some vaccines for JEV, but they are not widely available in Asia due to cost, licensing issues and safety concerns. JEV is mainly transmitted in the warm season, when large epidemics can occur. JEV most seen in temperate area of Asia. JE is primarily a children disease (0-14 year) but all age groups are affected. Approximately in JE cases 30% - 50% survivors have significant nuerologic disorder and 20% - 30% cases are fatal.

Capsid binds to viral RNA and forms a nucleocapsid that is enveloped by endoplasmic reticulam - derived membrane containing E and prM. E proteins are responsible for cellular attachment and possess a hydrophobic loop that mediates fusion of viral and host membranes. During JEV life cycle, the JEV virion undergoes a maturation and process that continuously fusion peptide from premature insertion into the cell membrane of the host. In an immature virion, E forms irregular timers with fusion loop capped by prM until it is cleaved in the trans-golgi network prior to viral secretion. E rearranges into an icosahedral network of flat antiparallel homodimers that bury the loop at their interface. Mature virion of JEV that attach to the cells and taken up into the endosome. This process penetrate the endosome and drags to the host and viral membranes, thereby releasing the nucleocapsid in to the cell of flavivirus E structure, and the major contacts are between the fusion loop and domain I(DI_-DIII pocket, not at the central region. The small dimer may be organized of E proteins from the viruses in the JEV serocomplex that provides an effective atomic model for JEV, E protein within mature virions. The flavivirus neutralizing antibodies binds E and inhibits the several stages of entry process, including attachment and fusion.

2. Materials and Methods

Docking

Molecular docking is a research technique which predicts the optimal orientation of one molecule to a second to form a stable complex. Knowledge of the preferred orientation is based on the strength and binding affinity between two molecules.

Autodock

Autodock is one of the most worldwide preferable molecular docking software. Autodock is a command line tool. It predicts the small 3D molecules and gives the binding energy of two molecules.

Programs

- AutoDock for docking the receptor and ligand
- AutoGrid for grid formation.

Licensed Under Creative Commons Attribution CC BY

PyMOL
PyMol is computer software, a molecular visualization tool. PyMol is universally accessible for scientific research and educational use.

Lipinski's Rule of five of SCFBio
Lipinski's Rule of Five helps in distinguishing between the drugs and drugs like chemical structure.Lipinski's Rule of Five predicts the probability of success and failure of the drug molecules.The drugs molecule should obey more than two rules of the following:

- Molecular mass less than 500 Dalton.
- High lipophilicity(expressed as LogP less than 5).
- Less than 5 H-bond donors.
- Less than 10 H-bond acceptots.
- Molar refractivity should be between 40-130.

PDB
Structural data of large biological molecules, such as proteins and nucleic acids. The data, typically obtained by X-ray crystallography, NMR spectroscopy, or, increasingly, cryo-electron microscopy. The Protein Data Bank (PDB) is a crystallographic database for the three-dimensional molecular structure submitted by biologists and biochemists from around the world are freely accessible on the Internet via the websites wwPDB Worldwide Protein Data Bank. We were downloaded the protein 3P54 from PDB Database (i.e. Fig -1).

Database used
- PDB
- Pubmed
- Pubchem
- Lipinski's Rule of five

Tools used
- Pymol
- Autodock
- Discovery Studio4.0

Protein preparation
- Target identification (3p54) from literature
- Download structure from PDB database

Ligand preparation
- Download ligands 3d structure from pubchem Tab-1
- Convert ligand sdf format to pdb format by pymol

Autodock setup
- Docking of 3P54 with each ligands set up of total grid (blind dock).Analyzed the result and found the region on protein, where ligand goes many times. Analysed the result (binding energy, H-bondinteraction, interacting residues).

3. Result and Discussion

The docking approach results of the three compounds are shown in the Tab- 2 and the interaction position between the ligands and the receptor are shown in the Fig-2(a), Fig-2(b), Fig-2(c). In FGIN-1-27 there is no H-bond interactions formation found. LYS124:HZ2, TRP233:HE1 and LYS166:HN, LYS166HZ3 are the H-bonds interactions of Cilnidipine and Niclosamide with receptor 3P54 respectively.It can be seen more clearly in the tables below.

4. Conclusion

3P54 is a suitable target for drug discovery, because of neutralizing antibodies bind E and can inhibit several stages of the entry process, including attachment and fusion. The binding affinity, H-bond interaction with inhibitors and information about interacting residues may give a positive impact on future drug discovery and to fight against deadly Japanese encephalitis.

Figure 1: Chain A of 3p54 protein.

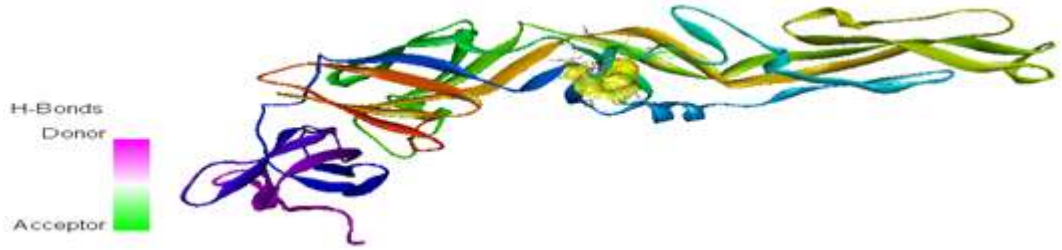


Figure 2(a): protein binding with inhibitor cilnidipine

Licensed Under Creative Commons Attribution CC BY

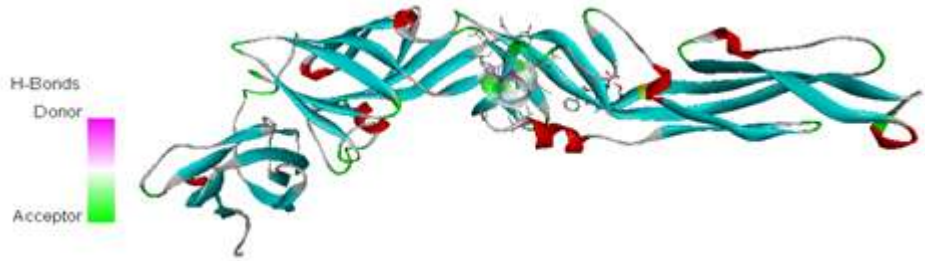


Figure 2(b): protein binding with inhibitor niclosamide

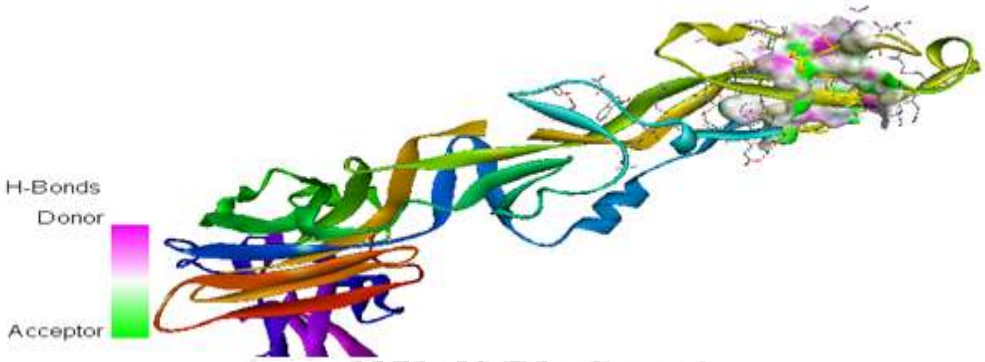


Figure 2(c): Protein binding with inhibitor FGIN-1-27.

Table 1

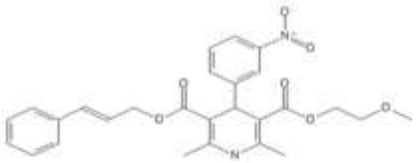
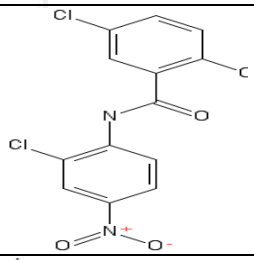
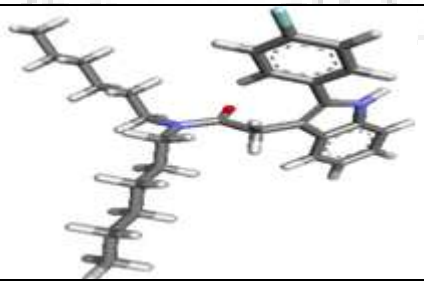
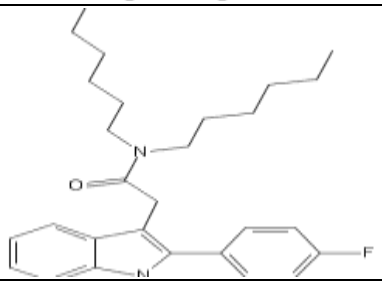
Sl No	Drugs	Information	3D structures	2D structures
1	cilnidipine	MW-492.528g/mol Compound 1D-5282138 Mf-$C_{27}H_{28}O_7$ XLogP3-4.275799 H-bond doner-1 H-bond acceptor-8 Rotatable bond-11 Molar Refractivity:133.643097		
2	niclosamide	MW-286g/mol Compound 1D-4477 Mf-$C_{13}H_8CL_2N_2O$ XLogP3-3.169539 H-bond doner-2 H-bond acceptor-5 Rotatable bond-2 Molar Refractivity:78.630379		
3	FGIN-1-27	MW-436g/mol Compound 1D-132496 Mf-$C_{28}H_{37}FN_2O$ XLogP3-7.505702 H-bond doner-1 H-bond acceptor-2 Rotatable bond-13 Molar Refractivity: 132.260696		

Table 2

Sl No	Drugs	Docking Score	H-bond Donner	H-bond acceptor	Torsional Energy	Atoms in H-bonds
1	cilnidipine	-4.9	1	8	3.58	LYS124:HZ2, TRP233:HE1
2	niclosamide	-5.43	2	4	1.19	LYS166:HN, LYS166HZ3
3	FGIN-1-27	-1.89	1	2	3.88	No H- bond formation

References

[1] Halstead SB, Jacobson J. Japanese encephalitis vaccines. In: Plotkin SA, Orenstein WA, Offit PA, editors. *Vaccines.* 5th ed. Philadelphia: Elsevier; 2008. pp. 311-52.

[2] Fischer M, Lindsey N, Staples JE, Hills S, Centers for Disease Control and Prevention (CDC). Japanese encephalitis vaccines: recommendations of the Advisory

Licensed Under Creative Commons Attribution CC BY

Committee on Immunization Practices (ACIP). *MMWR Recomm Rep* 2010; 59: 1-27 pmid: 20224546.
[3] Solomon, T. (2006). "Control of Japanese encephalitis – within our grasp?". *New England Journal of Medicine*. 355 (9): 869–71. doi:10.1056/NEJMp058263. PMID 16943399.
[4] Konno J, Endo K, Agatsuma H, Ishida N. 1966. Cyclic outbreaks of Japanese encephalitis among pigs and humans. Am. J. Epidemiol. 84:292–300
[5] Takahashi H, Pool V, Tsai TF, Chen RT. 2000. Adverse events after Japanese encephalitis vaccination: review of post-marketing surveillance data from Japan and the United States. Vaccine 18:2963–2969
[6] Xufang Y, Huanyu W, Shihong F, Xiaoyan G, Shuye Z, Chunting L, et al., et al. Etiological spectrum of clinically diagnosed Japanese encephalitis cases reported in Guizhou Province, China, in 2006. J Clin Microbiol 2010; 48: 1343-9 doi: 10.1128/JCM.01009-09
[7] Allison SL, Schalich J, Stiasny K, Mandl CW, Heinz FX. 2001. Mutational evidence for an internal fusion peptide in flavivirus envelope protein E. J. Virol. 75:4268–4275
[8] Chu JJ-H, Ng M-L. 2004. Interaction of West Nile virus with αvβ3 integrin mediates virus entry into cells. J. Biol. Chem. 279:54533–54541
[9] Davis CW, et al. 2006. West Nile virus discriminates between DC-SIGN and DC-SIGNR for cellular attachment and infection. J. Virol. 80:1290–1301
[10] Huang CY-H, et al. 2010. The dengue virus type 2 envelope protein fusion peptide is essential for membrane fusion. Virology 396:305–315
[11] Bressanelli S, et al. 2004. Structure of a flavivirus envelope glycoprotein in its low-pH-induced membrane fusion conformation. EMBO J. 23:728–738
[12] Modis Y, Ogata S, Clements D, Harrison SC. 2004. Structure of the dengue virus envelope protein after membrane fusion. Nature 427:313–319
[13] Nayak V, et al. 2009. Crystal structure of dengue virus type 1 envelope protein in the postfusion conformation and its implications for membrane fusion. J. Virol.83:4338–4344
[14] Kuhn RJ, et al. 2002. Structure of dengue virus: implications for flavivirus organization, maturation, and fusion. Cell 108:717–725
[15] Rey FA, Heinz FX, Mandl C, Kunz C, Harrison SC. 1995. The envelope glycoprotein from tick-borne encephalitis virus at 2 Å resolution. Nature 375:291–298
[16] Elshuber S, Allison SL, Heinz FX, Mandl CW. 2003. Cleavage of protein prM is necessary for infection of BHK-21 cells by tick-borne encephalitis virus. J. Gen. Virol. 84:183–191
[17] Stadler K, Allison S, Schalich J, Heinz F. 1997. Proteolytic activation of tick-borne encephalitis virus by furin. J. Virol. 71:8475–8481
[18] He RT, et al. 1995. Antibodies that block virus attachment to Vero cells are a major component of the human neutralizing antibody response against dengue virus type 2. J. Med. Virol. 45:451–461
[19] Oliphant T, et al. 2006. Antibody recognition and neutralization determinants on domains I and II of West Nile virus envelope protein. J. Virol. 80:12149–12159
[20] Thompson BS, et al. 2009. A therapeutic antibody against west nile virus neutralizes infection by blocking fusion within endosomes. PLoS Pathog.5:e1000453.
[21] Wu K-P, et al. 2003. Structural basis of a flavivirus recognized by its neutralizing antibody: solution structure of the domain III of the Japanese encephalitis virus envelope protein. J. Biol. Chem. 278:46007–46013
[22] Trott, O.; Olson, A.J. (2010), "AutoDock Vina: Improving the speed and accuracy of docking with a new scoring function, efficient optimization, and multithreading", *Journal of Computational Chemistry*, doi:10.1002/jcc.21334
[23] "PyMOL Molecular Graphics System". SourceForge..
[24] http://www.rcsb.org/
[25] Crystal Structure of the Japanese Encephalitis Virus Envelope Protein, strain SA-14-14-2.DOI: 10.2210/pdb3p54/pdb
[26] "PubMed: MEDLINE Retrieval on the World Wide Web". *Fact Sheet*. United States National Library of Medicine. 2002-06-07. Retrieved 2011-03-22.
[27] Roberts RJ (2001). "PubMed Central: The GenBank of the published literature". *Proceedings of the National Academy of Sciences*. 98 (2): 381–382. Bibcode:2001PNAS...98..381R. doi:10.1073/pnas.98.2.381. PMC 33354 . PMID 11209037.
[28] McEntyre JR; Ananiadou S; Andrews S; Black WJ; et al. (2010). "UKPMC: A full text article resource for the life sciences". *Nucleic Acids Research*. 39 (Database issue): D58–D65. doi:10.1093/nar/gkq1063. PMC 3013671. PMID 21062818.
[29] "PubChem Compound Results". *The PubChem Project*. USA: National Center for Biotechnology Information. Retrieved 7 January 2011.

Licensed Under Creative Commons Attribution CC BY