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Controlling Dengue- A review of battery of targets

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Abstract

Dengue fever occurs by Infection of mosquito-borne dengue virus and sometimes fatal dengue hemorrhagic fever. Every year cases of dengue infections are increasing. This suggests that the virus is becoming more virulent and its transmission is expanding. Nevertheless, no effective treatment for dengue infection currently exists. This paper has focused several potential targets for antiviral drug development against flavivirus. We discuss the flavivirus genome, NS1, NS2, NS3, Envelope protein and NS5 MTase domain and NS5 RdRp domain as a potential drug targets.

Keywords: Flavivirus, Dengue fever, Aedesaegypti, Structural protein, RNA dependent RNA polymerase.

1. Introduction

Flaviviridaeae are coated viruses that possess positive strand RNA genome that have been systamatized into three genera, Hepacivirus, Pestivirus, and Flavivirus. [1,2]. Members of the Flavivirus genus, e.g., dengue virus (DENV), yellow fever virus(YFV), Japanese encephalitis virus (JEV), tick-borne encephalitis virus, and West Nile virus (WNV), has significant role in medical science as an important arthropod-borne pathogens afflicting humans.

Dengue fever is considered as an ancient disease. It was first recorded in Chinese encyclopedia of disease and symptoms published during the Chin Dynasty (265 to 420 AD) [3]. In ancient Chinese medical literatures it referred as the Water poison because its vector is a flying insect which breed in clean water. Modern science has now approved that dengue fever is a viral disease transmitted between human hosts by Aedesaegypti. Aedesaegypti plays a significant role in the outbreak of dengue fever because of its well adaptation in urban living environment [4].

Dengue virus (DENV) is one of the most important arboviral infections. It affects more than 100 countries in the tropical and sub-tropical regions of the world [5]. Gubler *et al* found that every year dengue affects 50 million people which lead to more than 25,000 deaths because two fifth of the world population lives in areas where there is a risk of dengue infection [6]. Sickness caused by dengue virus comprises undifferentiated fever, dengue fever (DF), dengue hemorrhagic fever (DHF), and dengue shock syndrome (DSS) [7, 8].

As reported by WHO's 1997 documentation DF is clinically characterized as an intense feverish disease with two or more symptoms of retroorbital irritation, myalgia, headache, arthralgia, rash, and so on. Manifestations of DF can retain 2–7 days. DHF is described by the following norms: persistent high fever, hemorrhage tendency, hemoconcentration (>20%), and platelet counts (<100,000) [9- 12]. This paper starts with flavivirus genome, replication and their targets. This paper is a review of drug target of dengue which encourages drug development efforts.

Positive strand RNA genome of dengue virus encodes three structural proteins (C-prM-E), and seven nonstructural (NS) proteins (NS1-NS2A-NS2B-NS3-NS4A-NS4B-NS5), as schematically shown in Figure 1.

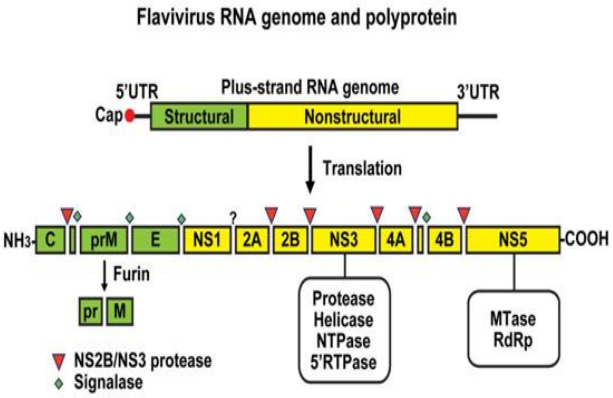

Figure 1: Above figure show organization of flavivirus genome and polyprotein processing

DENV genome contain 11 kb positive-sense, single-stranded RNA genome consist of a single open reading frame for 3 structural proteins (capsid (C), precursor membrane (prM) and envelope (E)) and 7 non-structural proteins (NS1-NS2A, NS2B, NS3, NS4A, NS4B, NS5). The open reading frame is bordered by untranslated regions. Sites of polyprotein cleavage mediated by the viral NS2B, NS3 and by host signalase and furin are shown, and the enzymatic activities of NS3 and NS5 are also indicated [13].

Entry of DENV in the host cell is occurred by receptor mediated endocytosis. Upon internalization and acidification of the endosome, fusion of viral and vesicular membranes allows release of the genomic RNA into the cytoplasm, which serves as mRNA. Translation of the single ORF at the rough ER produces a large polyprotein that is cleaved co- and posttranslationally into the mature proteins [14].

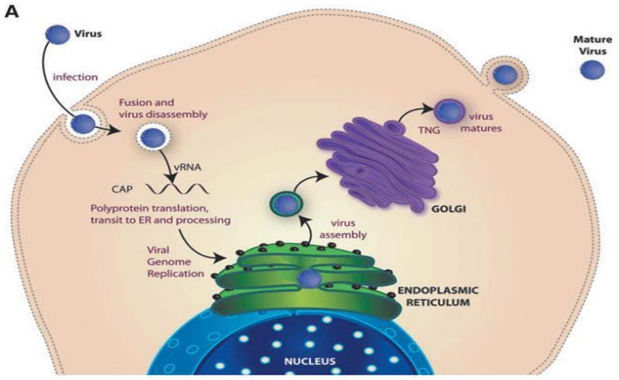

Figure 2: A typical life cycle after entering the host genome appears [14]

1.1 Drug targets for dengue

Dengue is a life threatening disease, No vaccine has been developed till now [15]. The main reason of this is presence of its four serotypes. It is compulsory to find a right drug target for dengue treatment so that an effective drug can be designed. Till now so much work has been done on many proteins of DENV. This review shall now concentrate all finding other potential targets for treatment.

NS2-NS3

NS3 is considered one of the most important nonstructural proteins because it has serine protease activity at its N terminal which cleaves the viral poly protein at its various regions. This function of NS3 is depends on its interaction with another nonstructural protein i.e. NS2B. Besides this NS2 also interact with other protein of DENV and these interactions play vital role in DENV life cycle so both NS2 & NS3 can be potential drug target. The table below now list the important landmark papers concerned with findings related to the protein.

Table 1: Authors finding on NS2-NS3

S. No.	Authors	Findings	Remarks
1.	Padmanaban Senthilvel *et al* [16]	They find the Flavonoid quercetin, found in papaya, with highest binding energy against NS2B-NS3 protease.	
2.	Rothan *et al.*[17]	They tested antiviral Activities in medicinal plants extracts against dengue virus using dengue NS2B-NS3 Protease assay.	
3.	Rothan *et al.*[18]	They worked on retrocyclin-1(RC-1) and reported that RC-1 reduces the rate of DENV replication and we may get recombinant RC-1 in *E.coli.* They considered recombinant RC-1 as a potent inhibitor.	
4.	Ayub *et al.*[19]	They analysed the conserved domains of NS3. They suggested that these domains of the NS3 sequence may be helpful in developing antiviral drugs.	One of the drawbacks with this target is that helicase activity of NS3 is not well understood.
5.	Muslum *et al.* [20]	They identified new allosteric site of NS2B-NS3 Protease. They utilized a define panel of just eight cysteine variants and only five cysteine reactive probes. They considered this site useful in the development of drug which allosterically inhibits NS2B-NS3 protease.	
6.	Leopoldo *et al.* [21]	They observed the importance of NS3 enhanced RNA- RNA interactions between molecules which is necessary for viral RNA synthesis. They proposed that, NS3 can regulate the folding or unfolding of viral RNA structures according to the ATP availability.	

NS5

DENV genome comprises about 11000 nucleotides in which NS5 is considered as a largest and most conserved NS protein. Its N terminal contains methyltransferase domain and C terminal contains RNA dependent RNA polymerase. Both domains have showed important role in viral replication. NS5 is also well characterized structurally and functionally. In future this target will show great contribution in the path of dengue drug development.

Table 2: Authors finding on NS5

S. No.	Authors	Findings	Remarks
1.	Hongping Dong *et al* [22]	They reported the methyltransferase activity of dengue virus (DENV) which sequentially methylates the guanine N-7 and ribose2′-O positions of viral RNA cap (GpppA→m7GpppA→m7GpppAm).	
2.	Lihui *et al* [23]	They demonstrated necessities of the N7 methylation activity for the WNV life cycle and, represented the methyltransferase as a novel and promising target for flavivirus treatment.	
3.	Christian *et al* [24]	They crystallized the RNA-dependent RNA polymerase (RdRp) domain of dengue virus serotype 3 (DENV-3) through a highly reproducible method. This method allowed structure refinement to a 1.79-Å resolution and revealed amino acids not seen previously. DENV-3 polymerase/inhibitor cocrystal structure at a 2.1-Å resolution was also conferred by them.	
4.	Egloff *et al* [25]	They worked on active MTase domain from DENV and represented their crystal structure. This structural information gives a unique ground for drug design against this emerging flavivirus.	

Envelope Protein

Envelope protein is made up of three domains involved in virus entry into host cell. It is well studied protein so it will be good option to find out inhibitor against Envelope protein.

Table 3: Authors finding on Envelope Protein

S. No.	Authors	Findings	Remarks
1.	Wahala *et al* [26]	They proposed that human antibodies directed to other epitopes on the virus are primarily responsible for DENV neutralization. Their results have implications for understanding protective immunity following natural DENV infection and for evaluating DENV vaccines.	
2.	Chiang *et al* [27]	They proposed the lapidated subunit vaccine for DEN serotypes and also other flavivirus members.	

Capsid Protein

It is structural proteins of DENV which interact with viral RNA and form nucleocapsid. By identifying inhibitors of this interaction we can devolve potential drugs.

Table 4: Authors finding on Capsid Protein

S. No.	Authors	Findings	Remarks
1.	Marcelo *et al* [28]	They found a link between lipid droplet metabolism and viral replication and suggested that change in this mechanism of lipid droplets could control viral replication.	

NS1 Protein

NS1 is non-structural glycoprotein. It is found on the cell surface and released into extracellular space**.** The flavivirus NS1 protein bears certain interesting and peculiar properties as compared to nonstructural proteins encoded by other RNA viruses this will make ideal target for drug development but structural biological information is very limited. This has hampered a complete understanding of its functions.

Table 5: Authors finding on NS1 Protein

S. No.	Authors	Findings	Remarks
1.	Yung-Chun Chuang, Shu-Ying Wang, Yee-Shin Lin, Hong- Ru Chen and Trai- Ming Yeh [29].	They did most of their work On NS1 and the antibodies and represented their role during Infection.	
2.	Muhammad Tahirul Qamar, Arooj Mumtaz, Rabbia Naseem, Amna Ali, Tabeer Fatima, Tehreem Jabbar, Zubair Ahmad & Usman Ali Ashfaq [30].	They focused on NS1 protein and concluded that flavonoids could serve as antiviral drugs for dengue infections.	

2. Conclusion

The purpose of this review is to point out several possible targets of dengue for antiviral drugs development. But, we feel NS5 may prove to be the target with the best chance of success.NS5 is the largest DENV protein, sharing a minimum of 67% amino acid sequence identity across the four DENV serotypes, NS5 is also the most conserved viral protein.

As per the target is concerned, NS5 can play the most appropriate role for the same. Being on the top of the list according to its size in different DENV serotypes it has sequence identity in 67% part of amino acids in all four DENV serotypes. RdRp domain of NS5 protein forms part of a multimeric complex that plays a crucial role in viral genome replication,

The development of new anti-dengue products from bioactive compounds is necessary in order to find more effective and less toxic anti-dengue drugs.

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Conflict of interest

There is no conflict of interest.

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