ARTHOPOD BORNE INFECTION ZIKA VIRUS

Geeta*1, Reshu Virmani2 Charan Singh3, Tarun Virmani3 and Jyoti Gupta3

1 CBS College of Pharmacy & Technology, Faridabad (Haryana).
2 Spectrum Institute of Pharmaceutical Sciences and Research, Gr. Noida (U.P).
3 School of Pharmaceutical Sciences, MVN University, Palwal (Haryana).

ABSTRACT

This review on zika virus is to provide the complete information about their vector and control the infection. Zika virus is an arthropod borne infection. Mosquitos are the main causative agent of this disease. Zika virus was first discovered in 1947 in Uganda in zika forest in a rhesus monkey. Zika virus (ZIKV; Genus Flavivirus, Family Flaviviridae) is an RNA-containing flavivirus transmitted in a zoonotic cycle between arboreal Aedes spp. mosquitoes and nonhuman primates in African and Asian forests. This virus is closely related to the other Flaviviridae of public health importance involving dengue, yellow fever, West Nile and Japanese encephalitis viruses. Transmission likely occurs through mosquito vectors from the Aedes genus of the Culicidae family in a sylvatic cycle including nonhuman primates, although antibodies have been found in a number of other mammals (i.e., water buffalo, elephants, zebras). Diagnostic tests for ZIKV infection include Polymerase Chain reactions (PCR) tests on acute-phase serum samples, which detect viral RNA, and other tests to detect specific antibody against ZIKV in serum. An enzyme-linked immune-sorbent assay (ELISA) has been discovered to detect immunoglobulin (Ig) M to ZIKV. The virus can be spread by sexually and by the bite of infected mosquitos and by other mammals. Vector control is the useful factor for control and prevention of this virus; vector should be controlled by using mosquitos repellent and by proper hygiene. There is no specific vaccine and medicine to treat zika virus infection, acetaminophen is used to relieve from pain and fever.

KEYWORDS: ZIKV, PCR, ELISA, Arthropods, Zoonotic, Aedes.
INTRODUCTION

Arthropod-borne viruses (arboviruses) cause significant human morbidity and mortality throughout the world.\textsuperscript{[1]} The first isolation of ZIKV was in 1947 from the blood of a sentinel Rhesus monkey No. 766, stationed in the Zika forest, near the Lake Victoria in Uganda, and in 1948 ZIKV was also isolated in the same forest from a pool of A. africanus mosquitoes.\textsuperscript{[2]} This spurt showcased the potential of ZF as an emerging disease, which could be misdiagnosed as dengue fever, as happened during the beginning of the Micronesian outbreak.\textsuperscript{[3]} Zika virus (ZIKV) is an RNA-containing flavivirus transmitted in a zoonotic cycle between arboreal Aedes spp. mosquitoes and nonhuman primates in African and Asian forests. The virus is nearly related to other flaviviruses of public health relevance including dengue (DENV), yellow fever and West Nile viruses, yellow fever, Japanese encephalitis viruses.\textsuperscript{[4]} These initial identifications were postdated by detection of ZIKV infection of humans, mosquitoes and animals in Africa and Asia by virus isolation and serological studies.\textsuperscript{[5]} A number of cases of infection with this virus have since been reported in Africa, India & Southeast Asia.\textsuperscript{[6]} Travelers might be a source of local transmission because Ae. albopictus mosquitoes are a suitable vector for ZIKV.\textsuperscript{[7]} In industrialized regions, where local transmission of arboviruses, such as dengue virus or chikungunya virus has been reported, physicians should test patients who comback from tropical regions for ZIKV when a case of dengue-like infection is doubtable.\textsuperscript{[8]} Although ZIKV has been known to circulate in both Africa and Asia since at least the 1950s, a bit is known about the genetic relationships between geographically distinct virus strains. Moreover, the geographic origin of the strains responsible for the epidemic that occurred on Yap Island, Federated States of Micronesia in 2007 and in 2010 pediatric case in Cambodia.\textsuperscript{[9]}

HISTORY OF ZIKA VIRUS

The first separation of ZIKV was in 1947 from the blood of a sentinel Rhesus monkey, close the Lake Victoria in Uganda, and in 1948 ZIKV was also separated in the same timberland from a pool of A. africanus mosquitoes.\textsuperscript{[2]} In duration of a series of studies of arboreal mosquitos as virus vectors in Uganda, 12 strains of Zika virus and one strain of another Group B arbovirus were separated between November 1961 and June 1963.\textsuperscript{[10]} The first well-documented report of human ZIKV infection was in Uganda in 1964 when Simpson depicted his own occupationally acquired disease.\textsuperscript{[9]} Serological and entomological data pointed ZIKV infections in the African landmass in Nigeria in 1971 and 1975, Sierra Leone in 1972 Gabon in 1975, Uganda in 1969 and 1970, Central African Republic in 1979, Senegal from 1988 to
1991 and Côte d'Ivoire in 1999. Until 2000, only few human cases were reported.\textsuperscript{[11]} April 2007, when a Zika fever (ZF) epidemic occurred in Yap Island in Micronesia, where 49 confirmed cases.\textsuperscript{[12]} Cambodia in 2010, during syndromic surveillance of patients with fever in neighboring Cambodia, a case of Zika virus infection in a young child was detected by PCR which pointed its presence in this area of Southeast Asia. A spurt in Yap Island, Micronesia in 2007, Micronesia leading to 99 cases.\textsuperscript{[14]} Lately in French Polynesia in 2013 and 2014 demonstrated the global allocation of this agent in Asia and the Pacific region, which in regards are the same regions where dengue is also indigenous. Different species of Aedes mosquitoes, including Aedes aegypti are the permissive vectors of Zika virus, and this mosquito species is also capable of transmitting dengue, making it creditable that Zika virus can also spread in areas in which dengue is indigenous.\textsuperscript{[13]} In the early 2015, several cases of patients showing symptoms of mild fever, rash, conjunctivitis and arthralgia were reported in the northeastern Brazil.\textsuperscript{[14]} ZIKV has the capability to spread to new locations where the Aedes mosquito vector is present and it could be a hazard for Southern Europe.\textsuperscript{[15]}

CAUSATIVE AGENT
It is a member of the genus \textit{Flavivirus} of the Family \textit{Flaviviridae}.\textsuperscript{[12]} Zika virus is an arthropod-borne \textit{Flavivirus} member of the Spondweni serocomplex, transmitted by \textit{Aedes} mosquitoes.\textsuperscript{[16]} The virus causes dengue-like syndromes such as rash, fever, arthralgia, headache and peri-orbital pain. To date, only Aedes mosquitoes have been known to transmit ZIKV. In Africa, the virus was isolated from both sylvatic and peri-domestic mosquitoes: Ae. Africanus, Ae. Apicocoargenteus, Ae. Luteocephalus, Ae. furcifer, Ae. vitattus and Ae. aegypti. In Asia, ZIKV was only isolated from a pool of Ae. aegypti caught from shop houses in the State of Pahang in Peninsular Malaysia.\textsuperscript{[6]}
VECTOR

Zika virus (ZIKV) is a mosquito-borne flavivirus from the Aedes genus (A. furcifer, A. taylori, A. luteocephalus and A. africanus) and monkeys\[1\], while humans are occasional hosts.\[12\] Transmission probably occurs via mosquito vectors from the Aedes genus of the Culicidae family in a sylvatic cycle involving nonhuman primates, although antibodies have been discovered in a number of other mammals (i.e., water buffalo, elephants, zebras). Shortly, Ae. aegypti is the only known vector to transmit the virus outside the African landmass, and Ae. albopictus has long been a suspected vector. Currently, Ae. albopictus has been shown capable of transmitting more than 20 arboviruses.\[2,21\] ZIKV was also isolated from Ae. Africanus and Ae. Apicoargenteus in Uganda and the Central African Republic from Ae. luteocephalus in Nigeria in 1969 and 1972 and from Ae. Vittatus, Ae. furcifer and Ae. aegypti in Cote d’Ivoire in 1999.\[9,15\] Spatially, the highest inoculation rates were conformed to the forest ground in June, forest ground and forest awning in September, forest awning in October and forest ground in November. Transmission was probably in December only in the forest.\[5, 17\]

Fig. 2: Life cycle of Aedes aegypti, there is an aquatic phase (larvae, pupae) and a terrestrial phase (eggs, adults).\[29\]

Fig. 3 Aedes Aegypti, one of the mosquito vector species for Zika virus.\[28\]
MODE OF TRANSMISSION

Further studies are needed to unveil whether co-infection and subsequent infection by different arboviruses can affect the course of the disease, the occurrence of severe cases and the ways of transmission (vertical, perinatal, sexual). Recent paper by Gourinat et al. (2015) shows evidence of virus secretion in urine for more than 10 days after onset of disease.\textsuperscript{[18]} Perinatal transmission and potential risk for transfusion-transmitted ZIKV infections has also been demonstrated. The virus was isolated from urine and semen of experimentally infected animals, and viremia developed in female boars that artificially inseminated with the infectious semen.\textsuperscript{[19]} Likewise, direct contact is also considered a potential route of transmission among humans, probably during sexual intercourse.\textsuperscript{[12]}

1. Sexual Transmission

Moreover, ZIKV transmission by sexual intercourse has been suggested by Foy et al., who described a patient who was infected with ZIKV in southeastern Senegal in 2008. Because the wife of the patient had not traveled out of the United States during the previous year and had sexual intercourse with him 1 day after he returned home, transmission by semen was suggested. Infectious organisms, especially sexually transmitted microorganisms including viruses (human papillomavirus or herpes simplex virus), are known to be etiologic agents of hematospermia. These results suggest that viral replication may have occurred in the genital tract, but we do not know when this replication started and how long it lasted.\textsuperscript{[12,22]}

2. Sylvatic Transmission

In 2008, a research program was initiated to investigate the mechanisms of sylvatic transmission of arboviruses in Kédougou, southeastern Senegal. The environmental factors that influence the abundance, distribution and infection of mosquito vectors that participate in the sylvatic cycles of several arboviruses were investigated beginning in June 2009. This species also entered in villages to feed on humans. Ae. furcifer was therefore considered to be the most important bridge vector between sylvatic CHIKV amplification and human populations.\textsuperscript{[5]}

INCUBATION PERIODS

Their study suggests that the extrinsic incubation period for ZIKV in mosquitoes is \( \approx 10 \) days.\textsuperscript{[15]} In a follow-up serum sample collected 36 days after symptom onset, IgG and IgM seroconversion against ZIKV was demonstrated.\textsuperscript{[20]}
CLINICAL FEATURES

Typically, clinical manifestations of human ZIKV disease are similar to many arboviral infections that betide without severe complications and may include a self-limiting feverish illness, arthralgia, myalgia, headache and maculopapular rash, Conjunctivitis, retro-orbital eye pain and lymphadenopathy have also been reported. Other symptoms involved sore throat, arthralgia, myalgia, rhinorhea and headache, elevated liver enzymes, which are presented in patients with acute dengue fever, are found in some, but not all, patients with Zika fever. [20,22,15,6,21]

![Rash caused by Zika fever.](image)

**Fig. 4** Rash caused by Zika fever. [28]

DIAGNOSIS

Diagnostic tests for ZIKV infection involve PCR tests on acute-phase serum samples, which find viral RNA, and other tests to discover specific antibody against ZIKV in serum. An ELISA has been evolved at the Arboviral.

**Virus Isolation**

The strains were gained from mosquitoes, humans and other mammals. Cultures supernatants were taken for virus RNA isolation. [27]

**RNA Extraction**

RNA was isolated from ZIKV stores. RNA was extracted in 50 µl of AVE buffer and stored at −80°C until use.
I. Recombination Detection
To help fill the gap in understanding ZIKV evolution,
(i) Adaptive genetic changes involving protein glycosylation design.
(ii) Phylogenetic relationship among separates and their spatiotemporal design of spread through Africa and Asia.
(iii) Dispersion through vertebrate reservoirs and invertebrate vector species.

The results pointed that ZIKV may have gone through recombination in nature and that, after it arised from Uganda in the early of the 20th century, it displaced to West Africa and Asia in the first half of the century, without any clear preference for host and vector species.[12] Zika virus, has a positive-sense, single-stranded RNA genome approximately 11 kilobases in length. The genome contains 5’ and 3’ untranslated regions joining a single open reading frame (ORF) that encodes a polyprotein that is cleaved into three structural proteins: the capsid (C), premembrane/membrane (prM), and envelope (E), and seven non-structural proteins (NS1, NS2A, NS2B, NS3, NS4A, 2K, NS4B, and NS5).[23]

II. Genetic and Phylogenetic Analyses
The E protein (≈53 kDa) is the major virion surface protein. E is involved in different aspects of the viral cycle, mediating binding and membrane fusion. The NS5 protein (≈103 kDa) is the largest viral protein whose C-terminal portion has RNA-dependent RNA polymerase (RdRP) activity and the N-terminus is included in RNA capping by virtue of its processing due to methyl transferase activity. The 3’NCR of the ZIKV genome contains about 428 nucleotides, including 27 folding patterns that may be involved in the recognition by cellular or viral factors, translation, genome stabilization, RNA packaging, or cyclization. Although diverse studies have contributed greatly to our understanding of the evolutionary biology of flaviviruses in general, few studies have addressed ZIKV evolutionary biology.[3]

III. Molecular Analysis
Acute and convalescent serum samples gathered 8 days apart from a 65 year old female were diagnosed at Forensic and Scientific Services (FSS), Queensland. Patient information provided on acute sample submission involved 2 days of illness with symptoms of rash, lethargy, nausea and joint pain and history of recent travel to the Cook Islands in March, 2014. Molecular assays involved two specific ZIKV real-time RT-PCRs patterned in the envelope (E) and nonstructural protein 1 (NS1) genes respectively.
ZIKV-E, Zika E For Primer: 5’-1222AAGTTTGCATGCTCCAAGAAAAT1244-3’,
Zika E Probe: 5’-FAM-1246ACCGGGAAGAGCATCCAGCCAGA1268-TAMRA-3’,
Zika E Rev Primer: 5’-1293CAGCATTATCCGGTACTCCAGAT1271-3’ and ZIKV-NS1,
Zika NS1 For Primer: 5’-3329GCACAATGCCCCACTGT3346-3’,
Zika NS1 Probe: 5’-FAM-3349TTCCGGGCTAAAGATGGCTGTTGT3373-TAMRA-3’,
Zika NS1 Rev Primer: 5’-3394TGGGCCTTATCTCCATTCCA3375-3’ (sequence positions based on ZIKV Yap 2007 GenBank accession number EU545988) using assay conditions as previously describe. A pan-flavivirus heminested RT-PCR targeting the non-structural protein 5 gene11 was also accomplished.

IV. Serological Analysis
Serological analyses pointed that the patient had previously been exposed to DENV, with detection of reactive flavivirus IgG and specific DENV-4 IgM antibodies in the acute sample. In contrast, seroconversions for both IgG and IgM were discovered between the paired acute and convalescent sera using the respective ZIKV IgG and IgM MIAs.[21]

V. The real-time RT-PCR assay analyzing mosquito and serum samples
Currently diagnosis of ZIKV infection is based on detection of specific antibodies or virus isolation from animals or mosquitoes which are time consuming. Standard RT-PCR and quantitative RT-PCR provide a rapid, specific and sensitive method for ZIKV early detection. However, real-time PCR, different to conventional assays, has many advantages, including rapidity, quantitative measurement, low contamination rate and easy standardization. One step rRT-PCR assay able to detect ZIKV strains circulating in Africa and Asia.[24,25]

VECTOR CONTROL
The epidemiology of ZIKV transmission on Yap Island appeared to be similar to that of dengue, strategies for prevention and control of ZIKV disease should involve promoting the use of insect repellent and intermediate to reduce the abundance of potential mosquito vectors. Officials responsible for public health surveillance in the Pacific region and keep in mind the possible diagnostic confusion between ZIKV illness and dengue.[15]

The mosquitoes convey large numbers of known and unknown viruses that infect humans, primates, mammals, birds, insects and plants.

The major objectives that need to be noticed if we are to win the war against these versatile viruses
• Develop vaccines to reduce the incidence of disease caused by known viruses;
• Develop therapeutic drugs to treat clinical diseases caused by known viruses;
• Develop unified vector-controlled strategies that will not unduly threaten the survival of wildlife species but locally will reduce the risk of disease in humans and animals.
• Develop universal teaching/training courses to be taught worldwide, to provide cores of expertise to implement these policies.
• Encourage the strengthening of levels of cooperation between academia and drug and vaccine development companies.
• Encourage the development of research programmes to understand the underlying mechanisms of arboviral pathogenicity, evolution, emergence and dispersal.
• Develop, at the international level, public health measures to inform and educate citizens in local arboviral disease control measures, including monitoring and reporting.
• Implement measures to improve monitoring procedures at borders, harbours, airports to reduce the influx of arthropods to new countries.
• Develop unified public health strategies for arboviral disease control.
• Simplify the procedures for establishing safety and efficacy of antiviral drugs, establish an international committee of experts charged with the objective of reviewing global arthropod control strategies.
• Develop and implement internationally acceptable and user-friendly guidelines for avoiding exposure to the different types of arthropod likely to carry human pathogens.

➢ The strategy on which to develop methods that will reduce the high morbidity and mortality rates due to human or animal arbovirus infections. Finally, we also have to recognise that arboviruses and arbovirus-related viruses infect more than just humans, invertebrates and land-based animals. They also infect plants, fish and marine animals. Let this be the mantra for arbovirus disease control in the future.[26]
Fig. 5 Typical water holding containers at individual homes including water barrels, coconut shells and cooking utensils.

TREATMENT
There is no vaccine to prevent or specific medicine to treat Zika infections.

- Get plenty of rest.
- Drink fluids to prevent dehydration.
- Take medicine such as acetaminophen to relieve fever and pain.
- Do not take aspirin and other non-steroidal anti-inflammatory drugs.
- If you are taking medicine for another medical condition, talk to your healthcare provider before taking additional medication.
- If you have Zika virus fever, prevent mosquito bites for the first week of your illness. During the first week of infection, Zika virus can be found in the blood and passed from an infected person to a mosquito through mosquito bites. An infected mosquito can then spread the virus to other people.\textsuperscript{[27]}
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