Zika Virus Infection: Damaging Consequences in Humans

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Abstract

An emerging arbovirus of the Flaviviridae family called ZIKV and others like dengue, West Nile, yellow fever, and Japanese encephalitis virus, causes a mosquito-borne disease transmitted by the Aedes genus, with recent outbreaks in the South Pacific. Here we examine the importance of human skin in the entry of ZIKV and its contribution to the induction of antiviral immune responses. Zika virus (ZIKV) is an arthropod-borne virus (arbovirus) in the genus *Flavivirus* and the family Flaviviridae. ZIKV was first isolated from a nonhuman primate in 1947 and from mosquitoes in 1948 in Africa, and ZIKV infections in humans were sporadic for half a century before emerging in the Pacific and the Americas. ZIKV is usually transmitted by the bite of infected mosquitoes. The clinical presentation of Zika fever is nonspecific and can be misdiagnosed as other infectious diseases, especially those due to arboviruses such as dengue and chikungunya. There is a high potential for ZIKV emergence in urban centers in the tropics that are infested with competent mosquito vectors such as *Aedes aegypti* and *Aedes albopictus*. Based on clinical criteria alone, ZIKV cannot be reliably distinguished from infections with other pathogens that cause an undifferentiated systemic febrile illness, including infections with two common arboviruses, dengue virus and chikungunya virus.

INTRODUCTION

Zika virus (ZIKV) was first isolated in 1947 from a rhesus monkey [1], and it was first described in a paper in 1952 though it was not given scientific attention. ZIKV was first described in Nigeria as reported by [2]. The first human ZIKV isolate came from a 10-year-old Nigerian female in 1954 [2]. ZIKV was isolated in mice inoculated with the patient's serum. Interpretation of the clinical presentation of the patient was difficult because the patient's blood also contained. ZIKV has long been known to occur in Africa and Southern Asia. About a decade ago about 15 cases of ZIKV has been described in scientific literature as reports of it came from yellow fever virus (YFV) serosurveys. 2007 was a year of ZIKV outbreak in Yap (an Island in the Western Pacific, Federated States of Micronesia) as reported by [3]. From there it migrated to Brazil in 2015 [4] and started spreading rapidly to other parts of South America, Central America, Mexico, and the Caribbean. ZIKV emerged in the Americas and, as in French Polynesia, appears to be associated with severe neurologic disease. An outbreak has also been reported in the Cape Verde Islands (Africa). It is believed that 1.5 million people have been infected since the outbreak in Brazil. The emergence of ZIKV in areas with cocirculation of other *Flavivirus*es will make diagnosis based on clinical and epidemiological grounds difficult and unreliable [5]. ZIKV was isolated in mosquito species collected during arbovirus studies in Africa and during fever studies in Asia [6-8]. ZIKV emergence was associated with the description of severe neurological complications: Guillain-Barré syndrome (GBS) in adults in French Polynesia and microcephaly in neonates in Brazil [9-11]. Dengue virus (DENV) and chikungunya virus (CHIKV) has been documented in French Polynesia [12, 13] and circulates mostly with ZIKV but most likely also occurs throughout the Americas, Asia, several Pacific islands, and Africa, where DENV and CHIKV are endemic. ZIKV is clearly following the routh of DENV and CHIKV, spreading to all countries infested with *Aedes aegypti* and *Aedes albopictus* mosquitoes [11].

Arboviruses: Important Studies

History

Arbovirus is an ecological term meaning contraction of arthropod-borne virus, hence defining viruses that are maintained in nature through biological transmission between a susceptible vertebrate host and a hematophagus arthropod like mosquito [14]. Arboviruses were first classified accord-

ing to serological criteria (antigenic classification) [15]. A new molecular basis for taxonomy is now used, and the genus *Flavivirus* is classified in clusters, species, and clades. The genus *Flavivirus* is composed of 53 virus species placed in three clusters: mosquito-borne viruses, tick-borne viruses, and viruses with no known vector (International Committee on Taxonomy of Viruses website chapter on virus families not assigned to an order, family Flavivirid). A fourth group of viruses found only in insects will also likely be placed in this genus [16]. For additional information on the history, definition, classification, taxonomy, and diagnosis of arboviruses, see previously published reviews [17].

Mode of Transmission

Most arboviruses cause zoonoses that usually depend on nonhuman animal species for maintenance in nature. Many animal species are host reservoirs (host of an infection in which the infectious agent multiplies and develops and on which the agent is dependent for survival in nature) of arboviruses; humans, with few exceptions (DENV, CHIKV, or YFV) are dead-end or accidental hosts (hosts from which infectious agents are not transmitted to other susceptible hosts). Other Aedes mosquito species (notably *Ae. africanus*, *Ae. albopictus*, *Ae. polynessiensis*, *Ae. unilineatus*, *Ae. vittatus* and *Ae. hensilli*) are considered as potential vectors of Zika virus. These species bite during the day. A vector of arboviruses may be defined as an arthropod that transmits the virus from one vertebrate to another by bite [18]. The most common mode of biological transmission is infection during a viremic blood meal and injection of infectious saliva during blood feeding (horizontal transmission). Nonvector arbovirus transmission has been reported to occur directly between vertebrates [19], from mother to child [20], nosocomially [21], by transfusion [22], via bone marrow or organ [23] transplantation, and sexually [24]. Given its epidemiology, the possibility of ZIKV transmission via transfusion should be considered as well [25]. To prevent potential ZIKV transmission by transfusion, a specific nucleic acid testing protocol was implemented during the French Polynesian ZIKV outbreak [26]. From November 2013 to February 2014, 42 (2.8%) of 1,505 blood donors tested were confirmed positive for ZIKV RNA; all of them were asymptomatic at the time of blood donation. Eleven of the 42 blood donors developed a "Zika fever-like syndrome" within 3 to 10 days after blood donation [27]. No transfusion-transmitted Zika fevers were documented during this outbreak, but the possibility that asymptomatic posttransfusion infection occurred cannot be ruled out. Unfortunately, blood samples collected within the first week after transfusion were not available. These results suggested that ZIKV can be transmitted by blood transfusion and that ZIKV nucleic acid testing can prevent the transmission of ZIKV by blood transfusion. ZIKV mode of transmission is represented in Fig 1.
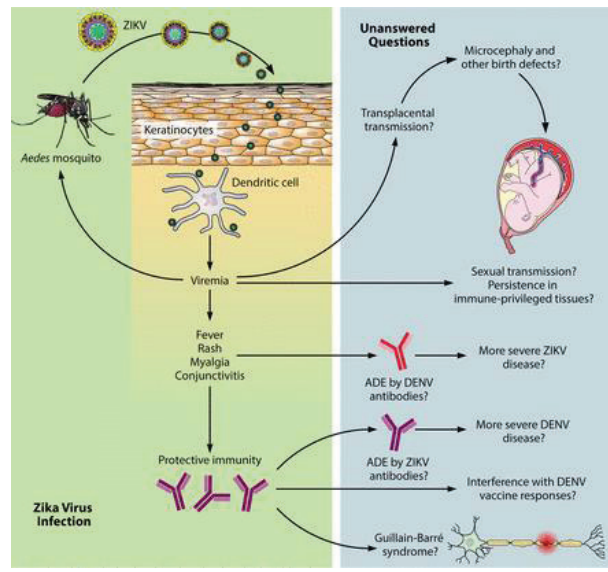

Figure 1: ZIKV Pathogenesis

The typical mode of transmission of ZIKV is represented in green background, with potential severe effects requiring further investigation represented in blue background. Dengue virus: DENV, antibody-dependent enhancement: ADE.

ZIKV SEROSURVEYS IN THE 1950S IN AFRICA AND ASIA

Interpretation of *Flavivirus* serological results is difficult because cross-reactions within this group of arboviruses were not well characterized when the first serosurveys were conducted. Discrepant results were observed when sera were tested by different methods [28] and even when the same method was used. Some studies reported results only for "arbovirus group B," but results for ZIKV were not available. ZIKV was not always included in the panel of antigens tested. Serosurveys for arboviruses were conducted by using a hemagglutination inhibition (HI) test, a CF test, an NT, a mouse protection test, a hemagglutination assay, and an enzyme-linked immunosorbent assay (ELISA). The HI test described by Clarke and Casals has been the most extensively employed. Nevertheless, although the data should be interpreted with caution, serosurveys suggest that ZIKV is endemic to Africa (East, Central, West, and South) and several countries in Asia. These global data were further confirmed by isolation of ZIKV from vectors and vertebrate hosts in most of these countries.

METHODS

Two scientists from America lived and worked in the village of Bandafassi in southeastern Senegal in August 2008 while

performing a mosquito-sampling project (on serosurveys of arboviruses) in surrounding villages. They later turned to be Patients 'A' and 'B' and they were men (36 and 27 years of age, respectively), and both had received the yellow fever 17D vaccine before their travel to Senegal. During their project, both patients reported being bitten often by wild Aedes spp. mosquitoes in the evenings while they worked. Both patients 'A' and 'B' left Bandafassi on August 21, stayed in Dakar for 2 days, and then returned to their homes in northern Colorado on August 24 and both became ill 6–9 days after their return. Symptoms in patient 'A' began on August 30 and consisted of swollen ankles, a maculopapular rash on his torso, and extreme fatigue and headache, but no fever was recorded. On August 31, he experienced the same symptoms and light-headedness and chills, wrist and ankle arthalgia, and symptoms of prostatitis (perineal pain and mild dysuria). However, he remained afebrile. Fatigue and rash decreased on September 1; only residual wrist arthralgia, headache, and prostatic symptoms persisted. On September 2, two aphthous ulcers appeared on his lip. On September 3, he and his wife observed signs of hematospermia (red–brown fluid in his ejaculate) that lasted until September 7. Patient 'B' experienced his symptoms during August 29–September 1, which included a maculopapular rash on his torso, extreme fatigue, headache, and swelling and arthralgia in his wrists, knees, and ankles. However, symptoms of prostatitis or hematospermia did not develop. Acute-phase blood samples were obtained from both patients on September 2.

In patient 'C' (a nurse and the wife of patient 'A') similar clinical symptoms developed on September 3, including malaise, chills, extreme headache, photophobia, and muscle pain that continued through September 6, without detectable fever. On September 7, a maculopapular rash developed on her torso that expanded to her neck and thighs on the following day, and an aphthous ulcer developed on her inside lip. On September 8, arthralgia in her wrists and thumbs and conjunctivitis developed. Her acute symptoms waned over the next several days. Patient 'C' had an acute-phase blood sample drawn on September 8. On September 11, she visited her primary care physician, who performed a complete blood count test and studies of hepatic function; all results were within reference ranges. Patient 'B' reported wrist arthralgia for 1 month after his acute illness, and patients 'A' and 'C' have had recurring wrist or thumb joint arthralgia since their acute illness. Convalescent-phase blood samples were drawn on September 22 from patients 'A' and 'N' and on September 26 from patient 'C.

Acute-phase and convalescent-phase paired serum specimens from the 3 patients were tested independently by several different laboratories. Results of virus isolation were negative for all samples when tested in vitro and *Aedes albopictus* mosquito (C6/36) cell cultures and by intracerebral inoculation of acute-phase serum of patient 'C' into suckling mice. Likewise, reverse transcription–PCRs with 16 different sets of arbovirus-specific primers did not detect arboviral RNA in any of the samples. Complement fixation tests against ZIKV and (Yellow fever virus) YFV antigens confirmed these interpretations. Hemagglutination inhibition, complement fixation, and virus neutralizing titers against ZIKV alone developed only in the convalescent-phase sample of patient 'C.

RESULTS

Unfortunately, the virus has two uncommon but severe complications that make it a menace to public health. The ZIKV outbreak in French Polynesia was associated with a twenty-fold increased risk of Guillain-Barré syndrome. This is an autoimmune disease, often triggered by infections, in which the immune system attacks the myelin lining of nerve cells, resulting in widespread weakness and paralysis. Weakness and paralysis spread from the legs upward. Two-thirds of patients lose the ability to walk, and 25% need to be put on a mechanical ventilator because of weakness of the respiratory muscles. Although most people make a partial or full recovery, 20% are still unable to walk at 6 months after diagnosis. The other condition linked to ZIKV is microcephaly, a birth defect in which a developing baby's brain fails to grow to its usual size. In Paraiba, one of the areas hardest hit by ZIKV, officials reported microcephaly in one out of every 100 newborns, a rate which is 100 times higher than usual. Hearing and vision problems have also been reported in newborns exposed to ZIKV in the womb. The vast majority of people with ZIKV are not very sick. In fact, most have no symptoms at all. In the outbreak on Yap, 77% of those with antibodies against Zika in their blood samples (indicating infection with the virus) were never ill.

In those who do get symptoms, the most common finding is an itchy red rash. Fever, headache, joint and muscle pains, and inflamed eyes are also frequent. People typically recover in 2 to 7 days, and death is rare. If a man were to get ZIKV through a mosquito bite, he could potentially infect his sexual partner and put their unborn baby at risk for microcephaly, a devastating neurological birth defect linked to the virus that doctors believe is caused by below-normal brain development in uterus. And since it's unclear how long Zika lasts in semen, and there is no commercial test available, it's not known how long a man needs to worry about possibly infecting his partner and his baby. Microcephaly causes lifelong physical and developmental problems for babies born with it. Problems can be wide-ranging, from seizures, to difficulty walking and learning, to hearing loss and vision difficulties. At birth, the pattern of brain damage he is seeing now looks distinct from microcephaly caused by other infections, such as cytomegalovirus (CMV) or rubella.

DISCUSSION

Vaccine development: Successful vaccination programs have reduced the global health burden of many *Flavivirus* infections. As no ZIKV vaccines have been tested even at the pre-clinical stage, we are likely years away from the introduction of a ZIKV vaccine. It is expected that at least some groups with existing *Flavivirus* vaccine platforms (e.g., chimeric live attenuated strains, pass aged or genetically engineered live attenuated strains, E protein subunit, subviral particles, inactivated virions, or DNA plasmid) will apply these strategies toward ZIKV vaccine development in an expedited manner. A major issue remains as to whether it will be easy or difficult to generate an immunogenic and safe vaccine against ZIKV. Given the relatively low variation between ZIKV strains (approximately 94% amino acid identity across the viral genome) and the lack of different genotypes or serotypes, it is

plausible that an effective vaccine against one strain would function broadly against all circulating ZIKV strains.

Development of therapeutics: Given that vaccines against ZIKV may be years away, the development of immediate measures to control or limit ZIKV disease should be a priority. To date, no drug screening studies have been published with ZIKV. Passive transfer or antibody-based therapeutics against ZIKV as prophylaxis or treatment may be possible, once strongly neutralizing human monoclonal antibodies are isolated, in analogy to studies performed with other Flaviviruses. Regardless of the approach, one obstacle to developing ZIKV therapeutics is that a key target population would be pregnant women; the design and implementation of trials to test new drugs in pregnant women will be challenging.

Animal models of ZIKV pathogenesis: Development of vaccines and therapeutics would be expedited by the development of animal models of the different manifestations of ZIKV diseases. There are few available data in nonhuman primates apart from the original isolation of ZIKV from the serum of a febrile rhesus monkey [1] and a study recently initiated to assess ZIKV infection dynamics in three rhesus macaques. There are also few available data in mice, as only three papers have reported on ZIKV infection in mice and nothing has been published in almost 40 years [29]. Although these studies suggested that ZIKV can replicate and cause injury in cells of the central nervous system, whether this pathogenesis is or is not related to the current linkages to GBS or microcephaly remains uncertain and requires further study. In addition to direct infection of newborn, juvenile, adult, and old mice, studies in which pregnant dams are inoculated with ZIKV and the effects on fecundity, neonatal infection, and brain development are evaluated could address the presumed linkage to microcephaly in humans.

Public health considerations: The association between ZIKV infection and neurological complications such as microcephaly prompted the World Health Organization to declare on 1 February 2016 a Public Health Emergency of International Concern surrounding the current ZIKV epidemic in Latin America and the Caribbean. The sudden surge of public health, clinical and basic science interest in ZIKV will increase our understanding of this virus that had remained an obscure viral curiosity until quite recently.

The lack of specific antiviral measures to combat ZIKV emphasizes the importance of vector control strategies for combatting arbovirus disease. Such approaches (removing sources of standing water that serve as breeding sites, larvicide and insecticide application, behavioral modifications to avoid mosquito exposure, and possibly the controlled introduction of genetically modified or sterile mosquitoes into an epidemic site) also will protect against DENV, CHIKV, and other mosquito-transmitted diseases [30].

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CONFLICTS OF INTEREST

The authors declare that there is no conflict of interest.

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