EDITORIAL

Microcephaly and Zika virus ☆

Microcefalia e vírus Zika

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The original isolation of the Zika virus (ZIKV), a *Flavivirus* member of the Flaviviridae family, was obtained in 1947 from the blood of a febrile rhesus monkey exposed at the Zika forest near Lake Victoria, in the outskirts of Entebbe, the capital of Uganda.[1] ZIKV was also isolated from wild mosquitoes in the same area and later periodic human febrile cases were attributed to ZIKV in Uganda and other countries in West and East Africa. Later, in the 1960s, ZIKV was detected in Asia and the virus was isolated from *Aedes aegypti* mosquitoes, initially in Malaysia and, subsequently, in several countries in Asia, showing that this arbovirus also occurred outside the African continent.[2] This new facet of ZIKV, *i.e.*, ability to cause epidemic disease transmitted by *Aedes aegypti*, disclosed a new milestone in the epidemiology of this arbovirus infection. It was clear that ZIKV had managed to adapt to an old acquaintance of humans, *Aedes aegypti* mosquitoes, transmitters of urban yellow fever, four serotypes of dengue fever, chikungunya virus, and other arboviruses in Asia and Africa.

Since the 1960s, sporadic cases of ZIKV infection have been reported in humans[3]; due to its sporadic occurrence

and low severity pattern, little importance was given to this arbovirus until a Zika fever epidemic occurred on Yap Island in the Republic of Micronesia in 2007, with the description of a rash febrile syndrome of mild intensity and a high percentage of asymptomatic cases.[4] This episode on Yap Island was followed by others, in the Pacific Ocean region of Polynesia and in some Southeast Asian countries, with outbreaks confirmed by serology or polymerase chain reaction (PCR) for the ZIKV on Easter Island, and in the Solomon Islands, the Cook Islands, Indonesia, Malaysia, Thailand, and French Polynesia.[5–7] In the latter, retrospective epidemiological studies suggested the occurrence of approximately 30,000 infections and, for the first time, cases of Guillain-Barre syndrome (GBS) were observed associated with ZIKV infection, as well as the notification of the first cases of perinatal transmission,[8] warning of the potential complications of congenital arbovirus infections, based on previous reports of encephalopathy, hemorrhagic fever, and fetal death, among others, associated with chikungunya and dengue viruses. In a retrospective analysis of live births during this outbreak in Polynesia, 17 cases of central nervous system malformations, including microcephaly in fetuses and newborns, were identified from March 2014 to May 2015. None of the pregnant women reported signs of ZIKV infection, but antibodies (IgG) to *Flavivirus* were found in four women tested by serology, suggesting asymptomatic infection.[9] Similarly as in Brazil, French Polynesia health authorities also believe

that ZIKV may be associated with birth defects if pregnant women are infected during the first or second trimester of pregnancy.

After the confirmation of the first cases of Zika fever in Brazil in May 2015, initially in the Northeast,[10] there was a rapid spread of the virus to other parts of the country, followed by the significant increase in notifications of newborns with microcephaly in the Brazilian Live Birth Information System (Sistema de Informação de Nascidos Vivos [SINASC]), with the recording of 141 suspected cases of microcephaly in November 2015 in the state of Pernambuco, after which an excessive number of cases in other northeastern states (Paraíba and Rio Grande do Norte) were detected, in addition to the records of miscarriages and stillbirths. Faced with this new scenario, the Ministry of Health of Brazil declared the event as a public health emergency of national concern.[11] It was also verified that the first months of pregnancy of children born with microcephaly matched the largest period of circulation of the ZIKV in the Northeast, and there was no correlation with family history of genetic disease, or tests demonstrating a pattern of other known infectious processes.

The causal association was carried out by Instituto Evandro Chagas (IEC) of the Ministry of Health through the isolation the ZIKV from brain tissue and detection of the virus in cerebral spinal fluid (CSF), brain tissue, and fragments of several viscera (heart, lungs, liver, spleen and kidney) of a newborn that died shortly after birth.[11] Subsequently, these results were reinforced with the detection of IgM antibodies to ZIKV in the CSF of 12 children born with microcephaly. All tests for other infectious agents associated with what is medically known as TORCH syndrome (toxoplasma gondii, other agents, rubella virus, cytomegalovirus, and herpes simplex virus, types 1 and 2), as well as dengue and chikungunya, were negative (Azevedo et al.; personal communication).

Another important contribution to elucidate the causal association was the identification of ZIKV the amniotic fluid of two pregnant women from the state of Paraiba with a history of rash illness and fetuses with microcephaly detected at the fetal ultrasonography.[12] After this finding, further studies were conducted, which allowed the complete sequencing of the virus isolated from the amniotic fluid, with the phylogenetic analysis disclosing that the virus shares 97-100% of its genomic identity with the Asian strain isolated during the outbreak in French Polynesia and that the presence of the viral genome in the patients for a few weeks after the acute phase suggests that the intrauterine viral load results from persistent replication.[13] As additional evidence, the identification of the ZIKV genome in placental cells in an 8-week miscarriage using RT-PCR techniques reinforced the potential of placental transmission.[14]

Recently, the Centers for Disease Control and Prevention (CDC) confirmed the presence of the virus, using RT-PCR and immunohistochemistry, in the brain tissue of four newborns with microcephaly and/or severe brain malformations that died after birth, and in the placentas of miscarried fetuses at 12 weeks of gestation.[15] Similar findings were identified by Mlakar et al.,[16] who identified the viral genome in the brain and placenta of a fetus miscarried in the 32nd week of gestation that had multiple brain lesions and intrauterine growth retardation detected after the 29th week of gestation,

which confirms the virus neurotropism, with a possible viral persistence in brain tissue and severe placental impairment.

Furthermore, there has been increasing evidence that in addition to the brain, the eyes would be the next target organ of ZIKV, as the presence of ocular disorders (macular atrophy) has been observed in children with microcephaly[17] and, more recently, macular and perimacular lesions with optic nerve atrophy,[18] as described in ten children with microcephaly during the Zika outbreak in the capital city of Salvador, state of Bahia.

Considering the severity of the situation, the rapid spread of ZIKV in the American continent, and the difficulties of diagnosis for an emerging arbovirus infection in the Americas, as well as the high risk of the virus spreading to other continents, the WHO declared the ZIKV epidemic an important international public health event, according to the International Health Regulations, and convened an emergency committee. The disclosure of the WHO note was followed by a description of the event.[19]

One of the major limitations to be overcome is the lack of commercial serological and molecular tests for the diagnosis of ZIKV, as the existing in-house tests are currently limited to reference laboratories, which are unable to meet the demands of public health laboratories. In fact, there is an urgent necessity to develop rapid tests (immunochromatographic), serological (IgM- and IgG-ELISA) and molecular tests for the early diagnosis of ZIKV infection, especially for the most vulnerable groups, i.e., pregnant women and individuals with autoimmune conditions and chronic diseases.[20] There have been recent records of deaths in patients with chronic diseases, lupus, hemolytic anemia, sickle-cell anemia, and others, which means these groups should have prioritized access to the diagnosis of ZIKV infection.

There is a clear need to reinforce antivectorial measures, which is the only currently available concrete measure to reduce cases of ZIKV infections. It is urgent that concrete actions be taken at all public levels together with the participation of society to reduce vector infestation indices; by reducing the number of vectors, we will reduce incidence rates and, obviously, the number of cases of microcephaly and other congenital malformations.

The Brazilian Ministry of Health made the political decision to develop a vaccine against ZIKV. Regarding this subject, there are several possible approaches to the development of a vaccine to prevent ZIKV infection, which include: inactivated virus vaccine, attenuated live virus vaccine, chimeric live virus vaccine, DNA vaccine, and subunit vaccine. Undoubtedly, the subunit and DNA vaccines are those that do not pose risks to pregnant women and special groups, and can be obtained quickly, the same occurring with the inactivated virus vaccine. It is important to note, however, that whatever the adopted formulation is, pre-clinical and clinical trials phases I, II, and III will take some years until a licensed product for use in humans is obtained.

In conclusion, the emergence of an ''almost unknown'' arbovirus and its causal association with different clinical manifestations and a degree of severity, especially observed in fetuses and adults with immune suppression and autoimmune disorders, opens up a range of opportunities for many studies, but the perspective of dissemination of this arbovirus to other continents in a globalized world will require a prompt response coordinated by the WHO,

together with the assistance and financial support of all member states and civil society worldwide.

Conflicts of interest

The authors declare no conflicts of interest.

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