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Chikungunya Virus (Chikv): General Characteristics and Possible Impact on Hemotherapy

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Abstract

Chikungunya virus (CHIKV) is an arbovirus belonging to the Alphavirus genus of the Togaviridae family. The virus is transmitted by Aedes mosquitos, the most important of which are A. aegypti and A. albopictus. CHIKV emerged in Africa but subsequent epidemics in the Indian Ocean led to viral dissemination in Asia. Although, some imported CHIKV cases were observed in various countries of the American continent, the virus demonstrated autochthonous transmission for the first time in the Caribbean island of Saint Martin in 2013. Consequently, CHIKV was rapidly spread to all Antilles and to continental America. In the majority of the cases, CHIKV causes abrupt illness with high fever and severe arthralgia. This virus causes explosive outbreaks with high morbidity and, in some cases, mortality. The initial phases of the infection are characterized by a high viral load and are without clinical symptoms. Therefore, such individuals can contaminate fresh blood products or hemo derivatives. The asymptomatic CHIKV infection, which occurs in ~ 25% of the infected individuals, can similarly be responsible for the transfusion transmission of this virus. Up to day, no measures exist to prevent the transmission of CHIKV by blood transfusion and deferral of blood donor candidates seems to be highly inefficient. The objective of this minireview is to summarize the current information on CHIKV general characteristics and epidemiology, with focus on blood transfusion.

Keywords: Arboviruses; Chikungunya virus; CHIKV; Viremia; Blood transfusion

Introduction

Chikungunya virus (CHIKV) is an arthropode-borne virus transmitted by Aedes mosquitoes (principally A. aegypti and A. albopictus). The virus was isolated between 1952 and 1953 from a Tanzanian febrile individual during a large outbreak of a dengue-like disease in the Southern part of the country [1]. Since then, CHIKV have been frequently documented as a cause of numerous outbreaks, which from Sub-Saharan Africa, were disseminated to the Southeast Asia. In 2013, CHIKV infection with autochthonous was reported in the Caribbean region of Central America and since then the virus has been rapidly spread to South and North America.

The disease caused by CHIKV is an acute illness characterized an abrupt onset with high fever (>38.9°C) and severe polyarthralgia. CHIKV incubation period, which is related to high viremic titers (105-1012 RNA copies/ml) ranges from 3 to 7 days and such individuals are able to contaminate fresh blood products and hemoderivatives. Moreover, asymptomatic CHIKV infections can occur in 5-25% of the cases [2]. Such characteristics of this arboviral infection additionally strengthen the hypothesis that this virus can be transmitted by blood transfusion. In this mini review we summarize the general properties of CHIKV and its possible impact on the transfusion of blood and hemoderivatives.

Taxonomy of CHIKV

CHIKV is an arbovirus transmitted by Aedes mosquitos and belongs to the Alphavirus genus of the Togaviridae family. The Alphavirus genus counts with 31 virus representatives, which are subdivided into two groups, according to the clinical symptoms, which they can cause in humans: New World viruses (causing encephalitis) and Old World viruses (causing polyarthritis/rash) [3,4]. CHIKV is a member of the Old World viruses group due its capacity to cause severe arthralgia, although it has been occasionally associated to atypical cases of neurological complications and hemorrhage [5]. Antigenically, CHIKV is a member of the Semliki Forest virus complex together with other alphaviruses like O'nyong-nyong, Mayaro and Ross River [3]. Although, antigenically and clinically CHIKV is most closely related to O'nyong-nyong virus (causes severe arthralgia), the genetic divergence between both viruses is significant [6].

Viral structure and replication

The CHIK virion is icosahedral and is enclosed by a lipid envelope of approximately 60-70 nm in diameter. Like the other alphaviruses, CHIKV has a positive sense, single-stranded RNA genome of approximately 11.8 kb and encodes two open reading frames (ORF), flanked by 5' and 3' untranslated regions (Figure 1) [7]. The first ORF, which spans two-thirds of the genome, is translated immediately into a polyprotein. It is subsequently cleaved into four non-structural proteins (nsP1-4), which have enzymatic properties and participate in the viral RNA replication. The second ORF (3'-ORF) is translated into 26S subgenomic RNA, also encodes a polyprotein which is processed to yield five structural proteins: capsid (C), envelope glycoproteins E1, E2 and E3, and a small 6K protein [8]. Glycoproteins E1 and E2 combine in a trimer form and are localized in the virion envelope. They participate during the initial stages of viral infection and mediate attachment and membrane fusion [9].

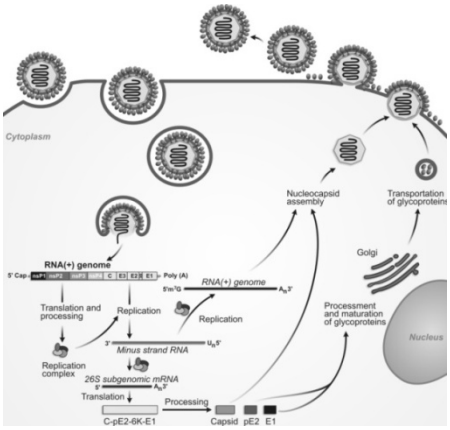

Figure 1: Replication cycle of Chikungunya virus (CHIKV) After the entry of the virus by endocytosis, the genome RNA of positive polarity is released in the cytoplasm. The organization of CHIKV genome is also demonstrated on the figure. The 5' -ORF is translated into non-structural proteins with enzymatic activities, which regulate the viral replication. After synthesis of a RNA strand with negative polarity, the viral cycle continues with its transcription into 26S subgenomic RNA which serves for the synthesis of the structural proteins (C, E1 and E2). At the same time the negative RNA is replicated into positive genomic RNA, which together with the structural proteins of the capsid forms the nucleocapsid. The envelope glycoproteins are glycosylated into the Golgi apparatus and are transported to the cell membrane. The nucleocapsid leaves the cell by budding incorporating its envelope with the glycoproteins.

The CHIKV replication cycle (Figure 1) is similar to the replication of the other alpha viruses [9]. It begins with the viral entry into the cell by endocytosis mediated via pH-dependent mechanism [10]. After formation of a fusion pore, the viral nucleocapsid is released into the cytosol. Glycoprotein E2 interacts with the cellular receptors, while E1 promotes viral fusion within the endosomes of the target cells. Following viral entry, the positive sense genomic RNA is released and the 5'-ORF is translated into the proteins nsP 1-4. These proteins assemble and form a viral replication complex, which is responsible for the synthesis of a negative-strand RNA intermediate. This RNA with negative-polarity is transcribed into sub genomic 26S RNA and genomic RNA, both of which participate in the translation of the structural proteins and in the assembly of new viral particles. The nucleocapsid assembly of CHIKV occurs in the cytoplasm by binding of the genomic RNA to the capsid. Virions leave the infected cell by budding through the cell membrane and simultaneously they acquire the envelope with the anchored glyco proteins [4].

Clinical manifestations

The infection by CHIKV starts with an asymptomatic incubation period, which lasts approximately 2 - 4 days [11]. Most of the infected individuals represent symptomatic infections; although serological surveys indicate that 3-25% of CHIKV-infected patients remain asymptomatic [12].

The classic clinical symptoms of the CHIKV infection are manifested by an abrupt onset of fever (generally with biphasic course of the temperature usually >38.9°C), incapacitating polyarthralgia, myalgia and sometimes, maculopapular rash [12]. The joint pains which involve the ankles and the knees, can be very intensive, and are usually symmetrical. Other joints can also be involved including these of the fingers, wrists, elbows, and shoulders. The arthralgic pains may last from weeks to months, and in some cases even years [5]. Other symptoms, resembling classical dengue infection include headache, asthenia, nausea and vomiting are often misdiagnosed. Generally, the incapacitating joint pains are used to make definitive diagnosis of the CHIKV infections. CHIKV infection is a self-limiting one and is non-fatal with acute phase which resolves within 3 to 7 days [12]. The chronic arthralgic phase, which is less severe, is characterized by fluctuations in the intensity of the joint pains and continuous relapses which can influence the quality of life [13]. Although, considered as a disease affecting the joints, CHIKV has been involved in some atypical symptoms described during epidemics like neurological complications, hemorrhage and death [5]. However, the atypical manifestations of CHIKV infection have been observed in children, especially in newborns, where comprise up to 53% of all cases [5].

The CHIKV induced viremia usually lasts from 5 to 7 days and the viral load in blood normally ranges from 105 to 109 viral RNA/ml but cases with lower viral load, especially in asymptomatic individuals have also been described [14]. The most important hematological abnormalities observed during CHIKV infection are pronounced lymphopenia and/or moderate thrombocytopenia and less commonly leukopenia [5].

Pathogenesis of CHIKV infection

Following mosquito bite and consequently intradermal viral spread, CHIKV enters the subcutaneous capillaries where its replication is initiated immediately in the dermal fibroblasts and macrophages [15]. CHIKV is disseminated through lymph node route and the blood stream to the liver, spleen, and the central nervous system, but studies in mouse models have shown that the virus targets preferentially muscles, joints and skin fibroblasts [16]. These cells appear to have a strict correlation to the pathogenesis and the clinical presentations of the CHIKV infection. Studies have shown that the disease symptoms are associated with presence of CHIKV in the muscles and joints and simultaneous infiltration of inflammatory cells in these compartments. In biopsies of infected mouse models, viral antigens were detected in fibroblasts of the joint capsule of the skeletal muscles and in the dermis [16,17]. Other studies in non-human primates and mouse models suggest that CHIKV replicates in high titers in joint tissues leading to the recruitment of inflammatory cells, especially monocytes, macrophages and natural killer cells [18,19]. Additionally, CHIKV infection in mice which lack $CD4^+$ T cells causes reduced joint swelling and less severe musculoskeletal tissue injury [20,21]. These findings demonstrate that the arthritic disease may be related to the interaction of CHIKV and the T cells [22].

CHIKV disease in humans is associated with elevated serum levels of specific cytokines and chemokines, including interleukine (IL)-1β, IL-6 and RANTES, which are related to more severe course of the CHIKV disease. Chronic arthralgia has also been associated with elevated levels of cytokines IL-6, granulocyte-macrophage colony-stimulating factor (GM-CSF) [23], mildly elevated C-reactive protein (CRP) and presence of CHIKV antigens and RNA in the synovial tissue [24]. Taken together, these data regarding the pathogenesis of CHIKV infection demonstrate an association between disease severity

and persistence of symptoms with the level of viral replication and the presence of inflammatory mediators at the site of infection.

Epidemiology of CHIKV infection

Epidemiologically, CHIKV has been circulating in Africa and Asia, causing extensive outbreaks. Two epidemiologic profiles of CHIKV transmission have been described: in Africa and in the Asian continent (Figure 2). In Africa, from where the virus originated, CHIKV is maintained in sylvatic cycles involving wild primates as reservoirs (and amplifying hosts) and forest dwelling Aëdes mosquitos as vectors (A. furcifer, A. vittatus, A. fulgens, A. luteocephalus, A. dalzieli, A. vigilax, A. camptorhynchites) which can also participate in the transmission of the yellow fever virus (YFV) [25]. During heavy rain falls, the sylvatic Aedes mosquitos can proliferate explosively, surpass the forest boundaries and introduce the infection in the nearby villages or urban centers (Fig.2). It is clearly demonstrated that A. furcifer, which is also the vector of YFV can enter in nearby villages, feed on humans and transmit CHIKV. For the passage of the sylvatic CHIKV infection in the urban zones, the participation of the highly domesticated mosquito A. aegypti has also been observed [25,26].

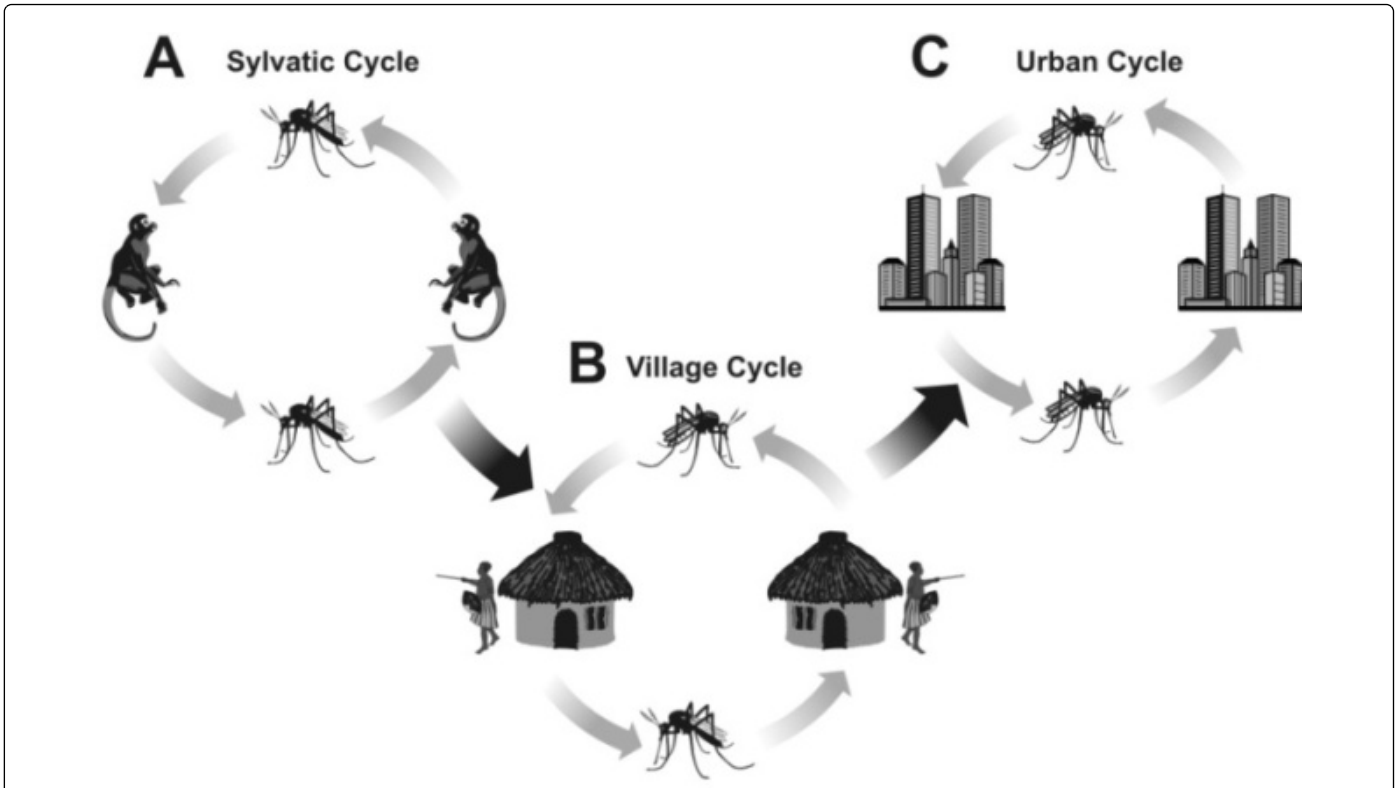

Figure 2: Epidemiologic cycles of Chikungunya virus. A. Sylvatic cycle. The virus is transmitted between non-human primates with the participation of many species of forest dwelling mosquitoes of the Aedes genus. B. Village cycle. During heavy rain falls, the forest dwelling mosquitoes enter in the nearby villages and can transmit the infection in humans. During this spread of the infection, the domesticated mosquitoes Aedes aegypti become infected and participate in CHIKV transmission. C. Urban cycle. The human migration towards large urban centers can translocate the infection, where the transmission is performed by the urban mosquitos A. aegypti/A. albopictus. The sylvatic cycle of CHIKV transmission has been described only in Africa, while in Asia the transmission is maintained only between humans (only urban cycle) without the participation of reservoirs. After the introduction of CHIKV in the Americas, the viral transmission is maintained exclusively in urban cycles.

On the other hand, in Asia, no animal reservoir has been identified for CHIKV and the transmission of the infection occurs only between humans. In Asia, the transmission of CHIKV is maintained by the urban dwelling A. aegypti; however, during the introduction of the virus from Africa, it acquired the A266V amino acid substitution in the E1 protein which increased its fitness to another mosquito vector: A. albopictus [27]. Therefore, in the urban centers of Asia, the virus is transmitted by both A. aegypti and A. albopictus but in the rural regions the transmission is maintained predominantly by A. albopictus [27,28]. Occasionally in the transmission of CHIKV have been involved other vectors like mosquitoes including Culex annulirostris, Mansonia uniformis and a wide variety of anopheles species [29].

The most important vector, however, participating actively in the urban cycles of CHIKV transmission during outbreaks remains A. aegypti. This mosquito is widely spread in the tropical regions of the world and over the 25 years there has been a global increase in its distribution [30]. Although, it demonstrates a typical tropical habitat, current reports describe its adaptation outside the tropics [31]. This probably depends on the fact that A. aegypti is well adapted on human blood feeding and can expand its areal outside the humid zones influenced by the human migrations. A. aegypti has day-biting activity in which the females feed on multiple hosts during their egg cycle, thus capable to transmit arboviral infections efficiently to many hosts. In Latin America, due to a massive program for vector elimination, as

imposed by the Pan American Health Organization, A. aegypti was eradicated in many countries by the early 1970s. However, due to the reduction of the control efforts, this mosquito re-infested, with exponential growth, many regions of Central and South America, and now it is the most important vector for the transmission of arboviral diseases in South America [31]

A.albopictus, or the so-called Asian tiger mosquito, due to its striped black white appearance, is an important vector of CHIKV in Southeast Asia, and in comparison to A. aegypti, which is strictly urban, A. albopictus is both urban and rural. This mosquito is natural of the Asian rainforests but it is well adapted also to urban localities. A. aegypti is relatively long-lived and lays very resistant eggs. The extreme resistance of the eggs is the reason which permitted the dissemination of A. albopictus to other regions of the world by exportation of egg-contaminated timber and tires from Southeast Asia. A. albopictus is aggressive, silent and similarly to A. aegypti, has a day-biting activity. Nevertheless, in contrast to A. aegypti, A. albopictus is both anthropophilic and zoophilic (i.e. feeds on humans and animals), which permits its participation in the dissemination of arboviral infections from animals to humans [31]. The introduction of A. albopictus in South America permitted its rapid spread on this continent due to the favorable climate conditions and now it is the second most important vector for the transmission of arboviral diseases in this continent.

CHIKV genotypes

The analysis of CHIKV sequences obtained from Africa and Asia demonstrate that the viral genomes have distinct genotypic characteristics and as a consequence, they are classified into three distinct genotypes (lineages). The genetic divergence between the CHIKV lineages is responsible also for their different antigenic properties. In Africa, circulate two genotypes designated as West African and East Central South African (ECSA) genotypes and they are responsible for the epidemics in this continent. In Asia, the epidemics are due to the Asian genotype. During the explosive CHIKV epidemic during 2005-2006 in the Indian Ocean, the ECSA genotype was introduced also in Asia, principally in India [32].

CHIKV and blood transfusion

The successful introduction and rapid spread of CHIKV in the Americas have been regarded as the most recent threat to blood safety in this locality. The transmission of the virus is extremely effective between the people, who remain highly viremic. The threat to blood transfusion is derived from the significant number of asymptomatic infections during epidemics and the high titers of CHIK viremia during the initial asymptomatic phase of the infection.

In a study performed during March-April in the Thepa and Chana districts of the Songkhla province in Thailand, 4.7% of the asymptomatic control group demonstrated CHIK viremia. During the follow up of these individuals, additional 4.7% seroconverted, giving a total prevalence of 9.4% for the CHIK viremia in asymptomatic individuals. The viral load ranged between 8.4x101 and 2.9x105pfu/mL, which were lower than the symptomatically infected patients with viral load between 1.3x101 and 2.9x108pfu/mL. The detected CHIKV RNA was probably a result of an initial phase of the infection as no anti-CHIKV IgM/IgG antibodies were confirmed [14]. The utility of the viral titer as a measure of symptomatic or asymptomatic infection cannot be applied, but even a minimal quantity of CHIKV RNA is probably able to contaminate blood derivatives as has been described for the dengue virus infection [33].

Similar situation was observed during the epidemic in the Reunion Island during 2005-2006, when after initial low number of cases, CHIKV caused explosive outbreak in February, 2006 affecting more than 25% of the whole island population. The isolated strains were very similar to the strains which caused the outbreak in the Democratic Republic of the Congo in 2000, and belonged to the ECSA Genotype, with clear divergence of the Asian strains [34]. During this outbreak, due to the extensive number of asymptomatic cases, the blood donation on the island was interrupted and the whole blood supply was imported from France. The peak risk of incidence was estimated to be during February when it was supposed that 1,500 donations per 100,000 (1.5%) will be found positive for CHIKV. However, this incidence could be much elevated due to the large number of positive cases [35].

The blood supply was interrupted also in Italy, when a focal CHIKV outbreak was registered in the Emilia-Romagna region in the northern part of the country [36]. Like the outbreak in the Reunion Island, this CHIKV circulation posed serious considerations to health authorities, regarding the effective control of the vectors and interruption of the blood donations which was related to a high cost. The emergence of CHIKV in the area of blood transfusion field, like the epidemics by West Nile virus (WNV) in the USA, demonstrates that virtually any arbovirus can cause initial asymptomatic viremia and can threat the blood transfusion safety. The emergence of WNV in the USA led to significant reorganization of the blood donation system, including implementation of WNV NAT for the routine testing of the blood donations [30].

In comparison with many chronically infecting viruses impacting blood transfusion like HIV, HCV and HBV, the arboviruses cause short-lived asymptomatic viremia and their circulation depends on arthropod vectors. Once the last are widely distributed in the tropical zones, the impact of the arboviruses in these regions is very significant. Moreover, they can cause explosive outbreaks with high morbidity, but at the same time a significant number of asymptomatic cases, responsible for the transfusion transmission of the respective arbovirus [30]. This scenario is currently complicated by our insufficient knowledge of the biology cycle and pathogenic impact of many arboviruses and the lack of suitable and highly specific diagnostic techniques for arboviral detection.

During the last decade, the CHIKV was uncommon in the Western Hemisphere. However, in 2013 the virus was successfully introduced in the Caribbean in the island of Saint Martin (French overseas territory) and rapidly spread to the majority of the Caribbean islands (Anguilla, Antigua and Barbuda, British Virgin Islands, Dominica, Dominican Republic, Guadeloupe, Haiti, Martinique, Puerto Rico, Saint Barthelemy, Saint Kitts and Nevis, Saint Lucia, Saint Martin, Saint Vincent and Grenadines, and Saint Maarten) (Figure 3) [37]. The Caribbean islands were a starting point from which CHIKV was disseminated to the inland South and North America (Brazil, Venezuela, Colombia, Ecuador, Central America, Mexico, and the USA) (Figure 3). By May, 2014 there were 103.018 suspected cases and more than 4.000 laboratory confirmed cases reported from the Caribbean region.

Figure 3: Distribution of Chikungunya virus infection in the Americas (source: Pan American Health Organization/World Health Organization. In: Data, Maps and Statistics, 2015).

Due to the wide spread of the vectors that transmit CHIKV on the American continent, it is possible that the virus can cause explosive outbreaks similarly to the observed in Africa and Southeast Asia. Nevertheless, CHIKV importance as a transfusion transmission threat in the Americas will depend on several factors: (1) prevalence of the CHIK viremia among blood donors and in the general population of the examined region; (2) the proportion of contaminated blood derivatives obtained from blood donors, which can infect transfused recipients; (3) clinical impact of the transfusion transmitted CHIKV infection in the respective receptor; (4) measures which are undertaken to reduce the transfusion transmitted CHIKV risk; and (5) the cost of the measures preventing transfusion transmission of CHIKV [38].

The effectiveness of blood donor deferral to prevent CHIKV transfusion transmission is unknown. The significant number symptomatic cases, which can surpass 75%, justifies such a process, nevertheless, the highest CHIKV load is detected in the initial stages of the viral infection and such individuals are asymptomatic [30]. They can easily contaminate blood derivatives used in hemotherapy. The remaining 25% of the infected individuals, which has unapparent infection, can similarly participate in this process. The interruption of the blood donations (like Reunion Island or Italy) seems to be highly effective in small areas but it is related to extreme cost for the blood supply and is not applicable for the most Latin American countries, which are highly populous and could not depend on outer sources of blood supply.

One possible measure to prevent the contamination of blood derivatives by CHIKV is to implement a highly sensitive PCR which can be used for viral detection in the blood donations [30]. However, the implementation of CHIKV NAT (nucleic acid amplification test) will depend on the seasonal emergence of epidemics and could result in high cost for the routine blood bank screening. Similarly to dengue, another arbovirus infection, where transfusion-transmitted cases during outbreaks have been reported in South America [33], CHIKV may significantly impact the blood donation process in this continent. Thus, implementation of CHIKV NAT can be a feasible solution during outbreaks, but it cannot be applied in the routine practice of blood donation even in the tropical countries where CHIKV is currently highly endemic.

Treatment strategies of the CHIKV induced diseases

Currently, there is no specific antiviral treatment of the CHIKV-induced diseases. However, there are some experimental strategies in development, which are supposed to influence the outcome of CHIKV infection. They include passive treatment with anti-CHIKV IgG antibodies, small synthetic molecules, and vaccines. The passive infusion of purified human anti-CHIKV IgG demonstrated high neutralizing effect in mice related to the therapeutic clearance of infection and protection of the immunized animals against lethal CHIKV infection course [39]. Although, passive anti-CHIKV IgG therapy has never been applied in humans, it could be helpful in neonates and immunocompromised patients, who demonstrate the highest possibility of severe CHIKV complications [39].

Other approaches including the use of small molecules, with synthetic or natural origin, have been experimentally tested for the treatment of CHIKV infection with varying efficacy. These molecules generally act as viral replication inhibitors by blocking the viral propagation at different stages but can also prevent viral entry into the cell. Some of these molecules include ribavirin, chloroquine, arbidol and a complex variety of natural compounds derived from plants.

Ribavirin is an established medicament, which is currently used for treatment of hepatitis C (HCV) [40] and respiratory syncytial (RSV) viral infections in humans [41]. Additionally, ribavirin has been experimentally applied for the treatment of a variety of infections including West Nile virus (WNV) [42] and SARS-corona virus [43]. It has been observed that the anti-WNV activity of ribavirin is based on error-prone replication [44]. The ribavirin capacity to treat arboviruses (in the case WNV) has served as a model and applied on CHIKV experimental infection. It has been observed that ribavirin can cause dose dependent reduction of the CHIKV load and can be more potent when applied in combination with interferon alpha (α)-2b [45]. Moreover, ribavirin in doses of 200 mg has been applied for the treatment of crippling arthritis in CHIKV-infected patients with improvement of the symptoms and reduction of the joint swelling [46]. Therefore, the ribavirin treatment should be considered for the treatment of CHIKV-induced complications.

Arbidol (ARB), another small molecule, is currently licensed in Russia and China for the treatment and prophylaxis of influenza A and B as well as other respiratory viral infections [47]. The derivates of ARB have demonstrated an inhibition effect on CHIKV infection and compared to the original ARB molecule, have much higher inhibition ratios [48].

Choloroquine, a well established drug to treat malaria, has been demonstrated to influence the in vitro inhibition of Sindbis and Semliki Forest alphaviruses by a mechanism, which up to date is unknown. The explosive outbreaks of CHIKV in the Indian Ocean, provided hope that this drug could be used similarly for the treatment of this arboviral infection. Although, in vitro studies have demonstrated clear chloroquine-inhibition of the proliferation of CHIKV-positive cells and the appearance of cythopathic effect, a randomized study performed in the Reunion Island (with explosive CHIKV epidemic) found no difference in the infection outcome between infected patients receiving placebo and chloroquine, thus not justifying the clinical use of this drug [49].

A large number of compounds derived from plants have been tested for its antiviral activity against CHIKV. Recently, harringtonine, a cephalotoxine alkaloid, was found to inhibit the CHIKV replication cycle, after viral entry into the cells. The successful use of this alkaloid

against Sindbis virus has suggested that harringtonine could inhibit the replication cycle of other alphaviruses [50]. Additionally, an alkaloid obtained from the green tea, epigallocatechin-3-gallate, was also found to inhibit the CHIKV infection in vitro. The mechanism of action of this compound is by blocking the CHIKV entry step into the cells, without effect on the viral replication [51].

Currently, similarly to the antiviral treatment of CHIKV there is no vaccine to prevent this infection. The first CHIKV vaccine was developed from formalin-inactivated virus grown on cultured monkey kidney tissue cells. It demonstrated excellent immunogenic response and no side effects were observed during its application in volunteers [52]. Another vaccine developed using the live attenuated CHIKV strain 15561 showed promising results, including decreased neurovirulence in newborn mice, reduced levels of viremia in monkeys and protection after further CHIKV infections [53]. This vaccine was well tolerated and safely applied during clinical trials, nevertheless, in phase II, mild arthralgias within the first 14 days after vaccination were observed in the CHIKV-vaccinated group [54].

The use of live-attenuated or inactivated virus for the development and clinical application of CHIKV vaccines had raised serious concerns for their safety, adverse effects and population coverage. To overcome these issues, the discovery of epitope-based vaccines against CHIKV infection is an increasing field as an alternative approach for vaccine design. The studies in this field use in silico approaches to discover immunogenic epitopes used to predict a peptide-based vaccine. Once this approach has been successfully applied for other viral infections, like rhinoviral [55], dengue [56] and Saint Louis encephalitis [57], its utilization for CHIKV immunotherapy seems quite promising.

Pratheek and coworkers [58] predicted that some CHIKV viral epitopes might activate, especially CD8+ T cell (CTL) response. The level of conservation of these epitopes and their binding affinities to the appropriate site of MHC-I/HLA-I were evaluated, including their CTL immunogenicity. Finally, it was demonstrated that two MHC-I (VLLPNVHTL and MTPERVTRL) e one HLA-I (FLTLFVNTL) epitopes are suitable for application in the development of epitope-based anti-CHIKV vaccine [58]. In another study, the peptide YYYELYPTM was found to be highly conserved among the different CHIKV strains and presents high and specific affinity for several HLA alleles. This demonstrates its capacity to activate the T cell-specific response. Furthermore, these studies also demonstrated that another peptide from the E2 protein (TPYEQKPGA) is more favorable to be used as a B cell epitope [59]. Taken together, these data establish potential candidates for a vaccine against CHIKV. Nevertheless, more efforts need to be implied in the development of both in vitro and in vivo studies to validate the epitopes affinity to the immunologic cells and their immunogenicity.

Conclusion

The entries of CHIKV infection in the Americas is a typical example of how arboviruses can surpass their natural zoonotic cycles and in a short time affect significant portions of the world. CHIKV successfully emerged in the Caribbean region and spread rapidly to the continental parts of North, Central and South America. The introduction of a completely unknown viral agent poses significant concerns in the infested region, including its impact and epidemic properties on a completely unprotected population and as consequence threat to the blood transfusion. Combining high titer asymptomatic viremia in the initial phases of the viral infection and significant percentage of asymptomatic infections as well as explosive outbreaks, CHIKV could negatively impact blood banks and cause significant economic loss. Therefore, the countries affected by CHIKV must establish rigorous policies for sensitive NAT CHIKV detection prompt diagnosis and even network communication measures to prevent viral transmission by transfusion of blood or hemoderivatives.

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