

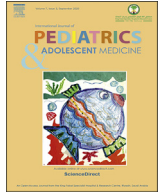
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Vertically transmitted chikungunya, Zika and dengue virus infections: The pathogenesis from mother to fetus and the implications of co-infections and vaccine development

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ABSTRACT

Chikungunya (CHIKV), Zika (ZIKV), and Dengue viruses (DENV) exhibit similar epidemiological and clinical patterns but have different pathophysiological mechanisms of disease manifestations. Differences occur in the severity of clinical presentations with the highest mortality in the general population attributed to DENV and neurological morbidity due to ZIKV. ZIKV and DENV infections can cause fetal loss with ZIKV exhibiting teratogenesis. CHIKV is associated with severe complications in the newborn. Co-circulation of the three viruses and the cross-reactive immune response between ZIKV and DENV viruses has implications for an attenuated clinical response and future vaccine development. Co-infections could increase due to the epidemiologic synergy, but there is limited evidence about the clinical effects, especially for the vulnerable newborn. The purpose of this paper is to review the pathophysiological basis for vertically transmission manifestations due to CHIKV, DENV, and ZIKV, to determine the potential effects of co-circulation on newborn outcomes and the potential for vaccine protection. Inflammatory cytokines are responsible for placental breaches in DENV and ZIKV; Hofbauer cells facilitate the transfer of ZIKV from the placenta to the fetal brain, and high viral loads and mechanical placental disruption facilitate the transmission of CHIKV. Co-infection of these viruses can present with severe manifestations, but the clinical and serologic evidence suggests that one virus predominates which may influence fetal transmission. All three viruses are in different stages of vaccine development with DENV vaccine being fully licensed. Antibody-enhanced infections in seronegative vaccinated candidates who develop natural infection to dengue limit its use and have implications for ZIKV vaccine development. Targeting transmission capacity in the vector could prevent transmission to all three viruses, and breast milk immunity could provide further clues for vaccine development.

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1. Introduction

Arboviral illnesses caused by the alphavirus Chikungunya (CHIKV) and flaviviruses Dengue (DENV) and Zika (ZIKV) have emerged as a global public health threat over the last decade. All three viruses exhibit similar ecological, biological and evolutionary patterns, and share common vectors, the *Aedes aegypti* and *Aedes albopictus* mosquitoes which have similar geographical distributions. These similarities result in epidemiological synergy [1] with

similar seasonality and attack rates. Sylvatic forms of *Ae. Aegypti* adapted to urban environments which facilitated global spread [2] and subsequently 215 countries or territories are suitable for the survival of both mosquitoes [3]. Combined with climate change, increased global transportation, deforestation in response to increased population density, human encroachment on wild habitats, and viral genome evolution, global distribution has been extensive and rapid in recent times [4–6]. Autochthonous vector-borne occurrences of CHIKV, DENV, and ZIKV, have been reported in 111,152 and 79 countries or territories respectively [7] with up to 123 countries or territories reporting more than one arboviral disease [3]. The three arboviruses have clinical similarities, exhibiting a short incubation period of 3–10 days and overlapping

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clinical features such as fever, maculopapular rash, myalgia, arthralgia, and retro-orbital pain. These similarities make a diagnosis on clinical grounds challenging in tropical areas where CHIKV, DENV, and ZIKV co-circulate [8]. Moreover, the high seroprevalence of DENV in areas of circulating ZIKV [9], their phylogenetic and antigenic similarities [10], and the cross-reactive immune response [11,12] complicates serologic approaches used to differentiate these two viruses. There is variability in the manifestations of clinical symptoms with CHIKV being symptomatic in 85% of cases [13], and ZIKV in 20% of cases [14] and up to 50% of cases in the Asian lineage [15] in contrast to most cases of DENV being asymptomatic [16]. Differences occur in the severity of manifestations with the highest mortality in the general population attributed to DENV and worst fetal outcome attributed to ZIKV [17–19]. In adults, complicated CHIKV presents with chronic joint pain, severe organ dysfunction and encephalitis in the elderly and persons with co-morbid conditions; DENV complications include hemorrhagic manifestations and plasma leakage leading to shock and death, and ZIKV is associated with Guillain–Barré Syndrome (GBS) [20]. Compared with CHIKV and ZIKV, DENV can be more severe in pregnant women than the general population with increased risks of oligohydramnios and antepartum and postpartum hemorrhage [21,22]. ZIKA and DENV infections can cause fetal loss and stillbirth with ZIKV exhibiting major teratogenic effects [23,24] and CHIKV with severe complications in the newborn [25]. The timing of infections and the severity of mother-to-child transmission differs among the arboviruses. ZIKV identified in the first, second, and third trimesters exhibit an 8%, 5%, and 4% risk of teratogenic birth defects, respectively [26], while a vertical transmission rate of 50% occurs in newborns infected intrapartum by CHIKV. Severe manifestations leading to death is rare [27,28].

Vector control is the primary method of control of arboviral illnesses but has faced challenges owing to insecticide resistance. The daytime biting pattern of *Ae. Aegypti* has rendered bed nets less effective, and finding and treating small larval breeding sites is also challenging [30]. No treatment is available for CHIKV, DENV ZIKV but precautions during travel to prevent mosquito bites and management is supportive care during illness. Novel antiviral therapies such as human CHIKV immunoglobulin are being studied to prevent mother-to-child CHIKV transmission in neonates born to viremic mothers and BCX4430, an adenosine nucleoside analogue with broad-spectrum antiviral properties against RNA viruses, is being investigated with ZIKV. Aziithromycin has also been shown to have antiviral properties against ZIKV [25].

Co-infections could increase due to the epidemiologic synergy and extensive global spread of these viruses, but clinical effects, especially for the vulnerable newborn remain largely unknown. The purpose of this paper is to review the pathophysiological basis for vertically transmission manifestations due to CHIKV, DENV, and ZIKV, to determine the potential effects of co-circulation on newborn outcomes and to review current vaccine strategies to prevent vertical transmission.

1.1. Host immunity

The innate immune response, primarily the type I interferon host response, is responsible for arbovirus infection control and neutralizing antibodies provide long lasting antibody-based immunity [15,29]. Following a bite, the infected mosquito inoculates the host dermis and epidermis with salivary contents, and the viral particles begin to infect target cells such as fibroblasts, monocytes, dendritic cells, and host endothelial cells [30–33]. During the inoculation, the mosquito saliva enhances viral replication and increases viral pathogenicity and viremia by attenuating the host response to the arbovirus [29,34–36].

Pregnancy does not confer increased susceptibility to arboviral illnesses except in the case of malaria where *Plasmodium falciparum* parasites selectively accumulate in the placenta [34]. Increased innate immunity, especially in the second and third trimesters provide an enhanced first response to infection to CHIKV, DENV, and ZIKV. The placenta of the pregnant host acts as an active immunological organ (innate immune system) capable of recognizing and responding to pathogens and is a potent immunoregulatory interface which protects the fetus from systemic infections. It undergoes periods of proinflammatory and inflammatory changes depending on the stage of gestation [35]. The first immunological phase is a proinflammatory phase that occurs during the first trimester. This phase facilitates implantation of the blastocyst in the endometrial tissue and trophoblastic replacement of the maternal blood vessels to ensure adequate fetal placental blood supply. The second immunological phase is an anti-inflammatory state during the period of fetal growth and development. In the final immunological phase, an influx of immune cells in the myometrium creates a proinflammatory environment to facilitate fetus delivery and placental expulsion.

Antiviral factors, secretory leukocyte protease inhibitor (SLP1) and interferon beta (IFN- β) are secreted by the trophoblast during the first trimester and are the first line of defense against viral infections. Most viral infections that affect the mother do not cause congenital fetal infection. Viral infections of the placenta stimulate the production of inflammatory cytokines such as tumor necrosis factor alpha (TNF α), interferon gamma (INF γ), and interleukins IL-12, and IL-6 which activate the maternal and fetal immune system. This inflammation results in placental damage, abortion, premature labor or fetal damage [25]. Despite the protective mechanisms outlined, genomic evolution of the arboviruses has conferred the ability to breach these protective barriers in different ways.

1.2. Chikungunya pathogenesis

The *Ae. Aegypti* saliva up-regulates anti-inflammatory genes such as IL-4 and IL-10 and downregulates inflammatory genes such as Toll-like receptor TLR-3, IL-2, IFN- γ , IFN- β , and TNF α during transmission of CHIKV [37,38]. CHIKV rapidly disseminates after inoculation by direct viral entry cell to cell transmission and infection of macrophages is facilitated when dying cells release apoptotic blebs. Manipulation and evasion of the host immune system facilitate viral replication [29]. CHIKV does not infect the placenta; therefore, incidences of miscarriages, stillbirths, premature delivery, and low birthweight newborns do not differ from uninfected pregnancies [37]. In vitro studies show that human syncytiotrophoblastic cell lines are refractory to infection [38]. Vertical transmission of 50% during the intrapartum period results in 12% symptomatic newborns, most of whom will develop severe manifestations of the disease [39]. The reason for this severe presentation is multifactorial and includes a high maternal viral load during the intrapartum period, chikungunya tropism for specific target organs, neonatal host factors, and the rate of cell division. Neonates have low levels of Toll-like receptor-induced interferon production, and this is necessary to prevent viral replication [28].

1.3. Dengue pathogenesis

During DENV transmission, the mosquito saliva enhances virus replication in human keratinocytes by suppressing the innate immune response [34,35]. Transmission also occurs by vertical transmission, via infected blood, organs or bone marrow.

When disease occurs, DENV effectively suppresses the IFN response. In patients with, nonsevere dengue, high levels of IFN α are found in the serum while those with severe dengue have low

levels of IFN α [30]. The primarily asymptomatic presentation in the general population may be related to the host's type 1 interferon and other immune response suppression of viral replication. In general, multifactorial host and viral factors can contribute to the severity of 4 serotypes of DENV. These include host genetic factors, secondary infection with a new serotype, activation of the complement system, a high viral load, and activation of nonprotective T cells leading to the increased plasma leakage characteristic of dengue hemorrhagic fever and dengue shock syndrome (DHF/DSS) [31]. Symptoms in the newborn could be very severe due to antibody dependent enhancement (ADE) if the maternal host had a secondary infection with a new serotype [31]. DENV infection in the pregnant host results in a placentitis which leads to hypoperfusion and subsequent fetal loss, prematurity, or low birth weight. Once the virus crosses the placenta during this inflammatory phase, developmental defects and neonatal infections can ensue [25].

1.4. Zika pathogenesis

Spontaneous mutation in the gene coding for Non-Structural Protein 1 (NS1) enhanced infectivity of the Asian lineage of ZIKV which may explain the 50% symptomatology compared with the African and South American lineages [7,14,15]. Innate immunity, ADE from previous flavivirus exposure and host immune response are key regulators of neuropathology. Similar to DENV, ZIKV can be transmitted through infected blood, organs or bone marrow as well as in saliva, urine, and semen. Similar to DENV a placentitis occurs during pregnancy which leads to hypoperfusion, fetal loss, and neonatal infections [25]. Additionally, placental macrophages or Hofbauer cells are permissive to ZIKA and facilitate viral transfer from the placenta to the fetal brain. Active replication of ZIKA in the placenta and fetal brain has been demonstrated in mice models and found in the placenta of women with fetal losses during the first and second trimester and in brain tissue of fetuses with microcephaly [40]. ZIKV is the only vertically transmitted flavivirus that can infect cortical progenitor cells.

1.5. Breast milk immunity

Concerning postpartum vertical transmission, both DENV and ZIKV particles have been found in breast milk, but studies are insufficient to conclude transmission via breast feeding [41,42]. Breast milk is known to increase white cells and TNF α levels during active infection in nursing infants, and immune regulation, modulation, and immune acceleration adapt to evolving organisms and infant requirements [43]. Breast milk provides an immunologic source that includes cytokines (IL-1, IL-8, transforming growth factor beta, and TNF) and chemokines (monocyte chemoattractant proteins MCP-1, MIP-1a, which regulate migration and infiltration of monocytes/macrophages and RANTES which is chemotactic for T cells, eosinophils, and basophils). These are in higher concentration than in the mother's general circulation and compensates for the infant's relatively immature immune system [44]. Thus far, there is no evidence to prove whether these elevated levels of chemokines and cytokines play a role in disease attenuation and hence, warrants further evaluation given the lack of infectivity through this route. The WHO continues to recommend long-term breastfeeding in mothers exposed to or infected with CHIKV, DENV, and ZIKV as the benefits outweigh the risks to the newborn.

1.6. Implications of co-circulation

The burden of arboviral illness and co-infection in pregnancy and fetal outcomes are mostly unknown [25]. Case reports and small cohort studies report increased disease severity with co-

infections. The evidence suggests that severe outcomes occur with co-infections but with similar frequencies as severe clinical cases with single arboviral infections [45]. It is uncertain how co-infections will interact with the fetoplacental immunity as DENV and ZIKV have similar pathogenic mechanisms that breach the placental barrier. Furthermore, ZIKV shares up to 59% sequence identity in the E protein with DENV resulting in antibodies induced against ZIKV E protein cross reacting with DENV E protein, thereby enhancing DENV replication and infection [46]. This cross-reactivity is compounded by an increased risk of severe presentations of ZIKV infection due to antibody-dependent enhancement (ADE) by a previous DENV infection [47]. Observational studies of co-infections suggest that clinical symptoms correlated with the virus that had a higher serum titer and that seroconversion occurred to one of two viruses at initial presentation. The impact on the fetus may be related to the predominant virus expressed, and severe reactions may occur by chance. Larger cohort studies are needed to evaluate enhanced disease severity.

1.7. Vaccine development

Based on the current understanding of fetal and neonatal outcomes, the burden of diseases, economic consequences and strain on the health care system, there is a need for vaccines, especially in low and middle-income countries. Some challenges limit development, but several vaccine candidates are undergoing evaluation.

1.8. Chikungunya

There are no licensed vaccines for CHIKV, but 15 vaccine candidates are currently under preclinical and clinical development. Logistical challenges in demonstrating efficacy in humans in phase II/III randomized controlled trials have been due to the unpredictable, focal and periodic nature of CHIKV outbreaks. Additionally, enrollment of mother-infant pairs limits phase II/III clinical trials for human immune intravenous immune globulin for neonates at risk for vertical transmission [48].

1.9. Dengue

The dengue vaccine, chimeric yellow fever virus-DENV tetra-valent dengue vaccine CYD-TDV (Dengvaxia[®]) has been licensed, and six other vaccines are under development [49]. The vaccine is efficacious in seropositive candidates, but seronegative vaccinated people developed severe presentations after natural infection. WHO guidelines recommend the determination of serostatus before vaccine administration. However, this strategy along with the absence of accurate, rapid diagnostic testing limits its cost-effective and safe use. Furthermore use in pregnancy is not recommended [50].

1.10. Zika

There are at least nine vaccine candidates for the Zika vaccine under clinical evaluation and 25 in nonclinical development to date. The Target Product Profile outlined by the World Health Organisation for vaccine use in an outbreak response would target persons aged nine years and over with 80% coverage of the population in order to prevent infection of the fetus up to one year after completion of the primary series [51]. Initial challenges to vaccine development are concerns that a ZIKA vaccine could result in an autoimmune trigger of GBS [20]. Animal studies have provided clues which suggest that neutralizing antibodies provide immune protection and sufficient titers protect against infection in the reproductive system, but this requires confirmation in clinical

Table 1
CHIKUNGUNYA, DENGUE AND ZIKA VIRUSES Geographical distribution, clinical presentation, pathogenesis of vertical transmission, AND vaccine and therapeutic developments.

	Chikungunya	Dengue	Zika
Distribution Countries/ territories	111	152	79
NonVector transmission route	Vertical transmission	Vertical transmission Infected blood, organs or bone marrow	Vertical Transmission Blood transfusion, saliva, urine and bone marrow, organ transplantation and semen
Clinical presentation	85% symptomatic	Primarily Asymptomatic	20% symptomatic overall 50% symptomatic (Asian Lineage)
Clinical complications General Population	Chronic joint pain, Severe organ dysfunction and Encephalitis (esp. In elderly and persons with comorbid illness)	Pleural effusion and Ascites Dengue hemorrhagic fever Dengue Shock syndrome Acute liver failure Myocarditis Encephalitis Kidney failure Death	Guillain–Barré Syndrome
Pregnancy	No increased risk in pregnancy	Oligohydramnios Antepartum hemorrhage Postpartum hemorrhage fetal loss; prematurity; low birth weight; developmental defects and neonatal infections	No increased risk in pregnancy
Pregnancy outcomes	50% vertical transmission during the intrapartum period results in 12% symptomatic newborns; most will develop severe multi-organ disease	Placentalitis and hypoperfusion	Fetal loss, developmental defects including microcephaly; optic nerve abnormalities; bilateral macular, and perimacular optic lesions; infant hearing loss
Pathogenesis of vertical transmission	A high maternal viral load during the intrapartum period and breaches in the placenta facilitate transmission	Placentalitis and hypoperfusion	Placentalitis; placental macrophages or Hofbauer cells facilitate viral transfer from the placenta to the fetal brain
Therapeutic modalities under trial	Chikungunya human intravenous immune globulin for at risk neonates		Short term monoclonal antibodies during pregnancy
Vaccine development	Vaccines under pre-clinical and clinical development	CYD-TDV (Dengvaxia [®] not recommended in pregnancy	Animal studies demonstrate immune protection from ZIKA in the reproductive system

trials. An additional strategy for pregnant women is the short term use of monoclonal antibodies which provides immunity as they await protective immunity from active immunization [52].

1.11. Vaccines and co-infections

Mice models demonstrate that immunization with inactivated DENV enhanced ZIKV infection, which has potential implications for maternal-fetal transmission and neurologic disease. Furthermore, inactivated ZIKV though highly protective against ZIKV challenged mice resulted in severe disease in DENV infected mice. These preliminary findings highlight safety considerations in the design and development of vaccines to be used in areas with cocirculating arboviruses [53]. Technology has attempted to interrupt transmission by targeting transmission capacity in the vector [32]. Vaccines that prevent the pathogen from completing its life-cycle in the vector are called transmission-blocking vaccines. Additionally, vaccines to mosquito saliva have the potential to target co-infections [33].

2. Conclusion

DENV, CHIV, and ZIKV are often found in co-circulation and share similar epidemiological patterns, clinical presentations, and cross-reactive immune responses, which highlight the importance of diagnostic vigilance. Though the pregnant state and the placenta protect the fetus, genomic evolution of the virus has found ways to breach these protective barriers in different ways through the modulation of the immune system. Similarities of transmission occur at inoculation of the pregnant host, but modification of the host innate immune system by the virus determines symptomatic presentation. Host and viral factors determine disease severity and pregnancy outcomes are determined by the timing of exposure to the fetus and ability to breach the placental barrier. Limited studies on outcomes with co-infections suggest severe disease occur by chance and that there is clinical and serological evidence of a predominant viral expression. Animal studies suggest the potential

to inhibit vertical transmission of CHIKV and ZIKV and limited transmission via breast-milk may reveal relevant information for future vaccine research (see Table 1).

Declaration of interest

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References

- [1] Bisanzio D, Dzul-Manzanilla F, Gomez-Dantés H, Pavia-Ruz N, Hladish TJ, Lenhart A, et al. Spatiotemporal coherence of dengue, chikungunya and Zika outbreaks in Merida, Mexico. *PLoS Neglected Trop Dis* 2018;12. <https://doi.org/10.1371/journal.pntd.0006298>. e0006298 PMID: 29543910.
- [2] Cao-Lormeau V. Tropical islands as new hubs for emerging arboviruses [letter]. *Emerg Infect Dis* 2016;22(5):913–5. <https://dx.doi.org/10.3201/eid2205.1505472>.
- [3] Leta S, Beyene TJ, De Clercq EM, Amenu K, Kraemer MUG, Revie CW. Global risk mapping for major diseases transmitted by *Aedes aegypti* and *Aedes albopictus*. *Int J Infect Dis* 2017;67:25–35. <https://doi.org/10.1016/j.ijid.2017.11.026>.
- [4] Gould E, Pettersson J, Higgs S, Charrel R, de Lamballerie X. Emerging arboviruses: why today? *One Health* 2017;4:1–13. <https://doi.org/10.1016/j.onehlt.2017.06.001>.
- [5] Gould EA, Higgs S. Impact of climate change and other factors on emerging arbovirus diseases. *Trans R Soc Trop Med Hyg* 2009;103:109–21.
- [6] Rezza G. Dengue and chikungunya: long-distance spread and outbreaks in naive areas. *Pathog Glob Health* 2014;108:349–55.
- [7] Sukhralia S, Verma M, Gopirajan S, Dhanaraj PS, Lal R, Mehla N, et al. From dengue to Zika: the widespread of mosquito-borne arboviruses. *Eur J Clin Microbiol Infect Dis* 2019;38:3. <https://doi.org/10.1007/s10096-018-3375-7>.
- [8] Paniz-Mondolfi A, Rodriguez-Morales A, Blohm G, Marquez M, Villamil-Gomez WE. ChikDenMaZika Syndrome: the challenge of diagnosing arboviral infections in the midst of concurrent epidemics. *Ann Clin Microbiol Antimicrob* 2016;15:42. <https://doi.org/10.1186/s12941-016-0157-x>.
- [9] Katzelnick LC, Coloma J, Harris E. Dengue: knowledge gaps, unmet needs, and research priorities. *Lancet Infect Dis* 2017;17. [https://doi.org/10.1016/S1473-3099\(16\)30473-X](https://doi.org/10.1016/S1473-3099(16)30473-X). e88–e100.
- [10] Barba-Spaeth G, Dejnirattisai W, Rouvinski A, Vaney MC, Medits I, Sharma A,

- et al. The structural basis of potent Zika-dengue virus antibody cross-neutralization. *Nature* 2016;536(7614):48–53.
- [11] Priyamvada L, Quicke KM, Hudson WH, Onlamo N, Sewatanon J, Edupuganti S, et al. Human antibody responses after dengue virus infection are highly cross-reactive to Zika virus. *Proc Natl Acad Sci USA* 2016;113:7852–7.
- [12] Bardina SV, Bunduc P, Tripathi S, Duehr J, Frere JJ, Brown JA, et al. Enhancement of Zika virus pathogenesis by preexisting antinflavivirus immunity. *Science* 2017;356:175–80.
- [13] Weaver SC, Lecuit M. Chikungunya virus and the global spread of a mosquito-borne disease. *N Engl J Med* 2015;372:1231–9.
- [14] Pawitwar Shashank S, Dhar Supurna, Tiwari Sneham, Chet Raj Ojha, Jessica Lapiere, Kyle Martins, et al. Overview on the Current Status of Zika Virus Pathogenesis and Animal Related Research. *J Neuroimmune Pharmacol* 2017;12:371–88. <https://doi.org/10.1007/s11481-017-9743-8>.
- [15] Gallian P, Cabie A, Richard P, Paturel L, Charrel RN, Pastorino B, et al. Zika virus in asymptomatic blood donors in Martinique. *Blood* 2017;129:263–6.
- [16] Schmit MA, Diamond MS, Harris E. Dendritic cells in dengue virus infection: targets of virus replication and mediators of immunity. *Front Immunol* 2014;17:647. <https://doi.org/10.3389/fimmu.2014.00647>.
- [17] Bodie-Williams SS, Knowles SO. Viral mosquito-borne illnesses in pregnancy: a look at Aedes. *Obstet Gynecol Int J* 2016;5. <https://doi.org/10.15406/ogij.2016.05.00139>. 00139.
- [18] Marinho PS, Cunha AJ, Amim Junior J, Prata-Barbosa A. A review of selected Arboviruses during pregnancy. *Mat Health, Neonatol Perinatol*. 2017;3:17. <https://doi.org/10.1186/s40748-017-0054-0>.
- [19] Basurko C, Carles G, Youssef M, Guindi WE. Maternal and fetal consequences of dengue fever during pregnancy. *Eur J Obstet Gynecol Reprod Biol* 2009;147:29–32.
- [20] Weaver SC, Charlier C, Vasilakis N, Lecuit M. Zika, chikungunya, and other emerging vector-borne viral diseases. *Annu Rev Med* 2017;69:395–408. <https://doi.org/10.1146/annurev-med-050715-105122>.
- [21] Ismail NA, Kampan N, Mahdy ZA, Jamil MA, Razi ZR. Dengue in pregnancy. *Southeast Asian J Trop Med Publ Health* 2006;37(4):681–3.
- [22] Machado CR, Machado ES, Rohloff RD, Azevedo M, Campos DP, de Oliveira RB, et al. Is pregnancy associated with severe dengue? A review of data from the Rio de Janeiro surveillance information system. *PLoS Neglected Trop Dis* 2013;7. <https://doi.org/10.1371/journal.pntd.0002217>. e2217.
- [23] Paixao ES, Teixeira MG, Costa MD, Rodrigues LC. Dengue during pregnancy and adverse fetal outcomes: a systematic review and meta-analysis. *Lancet Infect Dis* 2016;16:857–65.
- [24] Franca GV, Schuler-Faccini L, Oliveira WK, Henriques CM, Carmo EH, Pedi VD, et al. Congenital Zika virus syndrome in Brazil: a case series of the first 1501 live births with complete investigation. *Lancet* 2016;388:891–7.
- [25] Charlier C, Beaudoin MC, Couderc T, Lortholary O, Lecuit M. Arboviruses and pregnancy: maternal, foetal, and neonatal effects. *Lancet Child Adolesc Health* 2017;1:134–46.
- [26] Shapiro-Mendoza CK, Rice ME, Galang RR, Fulton AC, VanMaldeghem K, Prado MV, et al. Zika pregnancy and infant registries working group pregnancy outcomes after maternal Zika virus infection during pregnancy—U.S. Territories, January 1, 2016–April 25, 2017. *MMWR Morb Mortal Wkly Rep* 2017;66:615–21.
- [27] Gerardin P, Barau G, Michault A, Bintner M, Randrianaivo H, Choker G, et al. Multidisciplinary prospective study of mother-to-child chikungunya virus infections on the island of La Reunion. *PLoS Med* 2008;5. <https://doi.org/10.1371/journal.pmed.0050060>. e60.
- [28] Evans-Gilbert T. Chikungunya and neonatal immunity: fatal vertically transmitted chikungunya infection. *Am J Trop Med Hyg* 2017;96:913–5. <https://doi.org/10.4269/ajtmh.16-0491>.
- [29] Long KM, Heise MT. Protective and pathogenic response to chikungunya virus infection. *Curr Trop Med Rep* 2015;2:13–21. <https://doi.org/10.1007/s40475-015-0037-z>.
- [30] Schmid MA, Diamond MS, Harris E. Dendritic cells in dengue virus infection: targets of virus replication and mediators of immunity. *Front Immunol* 2014;5:647. <https://doi.org/10.3389/fimmu.2014.00647>.
- [31] Martina BEE, Koraka P, Osterhaus ADME. Dengue virus pathogenesis: an integrated view. *Clin Microbiol Rev* 2009;22(4):564–81. <https://doi.org/10.1128/CMR.00035-09>.
- [32] Bordi L, Caglioti C, Lalle E, Castilletti C, Capobianchi MR. Chikungunya and its interaction with the host cell. *Curr Trop Med Rep* 2015;2:22–9.
- [33] Hamel R, Dejarnac O, Wichit S, Ekcharyawat P, Neyret A, Luplertlop N, et al. Biology of Zika virus infection in human skin cells. *J Virol* 2015;89(17):8880–96. <https://doi.org/10.1128/JVI.00354-1>.
- [34] Wichit S, Ferraris P, Choumet V, Missé D. The effects of mosquito saliva on dengue virus infectivity in humans. *Curr Opin Virol* 2016;21:139–45. <https://doi.org/10.1016/j.coviro.2016.10.001>.
- [35] Green AM, Beatty PR, Hadjilaou A, Harris E. Innate immunity to dengue virus infection and subversion of antiviral response. *J Mol Biol* 2014;426:1148–60. <https://doi.org/10.1016/j.jmb.2013.11.023>.
- [36] Wichit S, Diop F, Hamel R, Talignani L, Ferraris P, Cornélie S, et al. Aedes Aegypti saliva enhances chikungunya virus replication in human skin fibroblasts via inhibition of the type I interferon signaling pathway. *Infect Genet Evol* 2017;55:68–70.
- [37] Fritel X, Catteau C, Calliez F, Brode A, Vaillant JY, Ansquin H. Chikungunya outbreak, pregnancy outcome and perinatal mortality: Observational study of about 40,000 pregnancies and deliveries on Réunion Island during 2004–2006. 13th international congress on infectious diseases Poster # 49.013. [doi:10.1016/j.ijid.2008.05.879](https://doi.org/10.1016/j.ijid.2008.05.879)
- [38] Couderc T, Chretien F, Schilte C, Disson O, Brigitte M, Guivel-Benhassine F, et al. A mouse model for chikungunya: young and inefficient type-1 interferon signaling are risk factors for severe disease. *PLoS Pathog* 2008;4:e29.
- [39] Lenglet Y, Barau G, Robillard PY, Randrianaivo H, Michault A, Bouveret A, et al. Chikungunya infection in pregnancy: evidence for intrauterine infection in pregnant women and vertical transmission in the parturient. Survey of the Reunion Island outbreak. *J Gynecol Obstet Biol Reprod (Paris)* 2006;35:578–83.
- [40] Bhatnagar J, Rabeneck DB, Martines RB, Reagan-Steiner S, Ermias Y, Estetter LBC, et al. Zika virus RNA replication and persistence in brain and placental tissue. *Emerg Infect Dis* 2017;23:405–14. <https://doi.org/10.3201/eid2303.161499>.
- [41] Colt S, Garcia-Casal MN, Pena-Rosas JP, Finkelstein JL, Rayco-Solon P, Weise Prinzo ZC, et al. Transmission of Zika virus through breast milk and other breastfeeding-related bodily-fluids: A systematic review. *PLoS Negl. Trop. Dis.* 2017;11. e0005528.
- [42] Mann TZ, Haddad LB, Williams TR, Hills SL, Read JS, Dee DL, et al. Breastmilk transmission of flaviviruses in the context of Zika virus. A systematic review. *Paediatr Perinat Epidemiol* 2018. <https://doi.org/10.1111/ppe.12478> [Epub ahead of print].
- [43] Riskin A, Almog M, Peri R, Halasz K, Srugo I, Kessel A. Changes in immunomodulatory constituents of human milk in response to active infection in the nursing infant. *Pediatr Res* 2012;71(2):220–5.
- [44] Labbok MH, Clark D, Goldman AS. Breastfeeding: maintaining an irreplaceable immunological resource. *Nat Rev Immunol* 2004;4:565–72.
- [45] Vogels CBF, Rückert C, Cavany SM, Perkins TA, Ebel GD, Grubaugh ND. Arbovirus coinfection and co-transmission: a neglected public health concern? *PLoS Biol* 2019;17. e3000130. <https://doi.org/10.1371/journal.pbio.3000130>.
- [46] George J, Valiant WG, Mattapallil MJ, Walker M, Huang YJS, Vanlandingham DL, et al. Prior exposure to Zika virus significantly enhances peak Dengue-2 viremia in Rhesus Macaques. *Sci Rep* 2017;7:10498. <https://doi.org/10.1038/s41598-017-10901-1>.
- [47] Bardina SV, Bunduc P, Tripathi S, Duehr J, Frere JJ, Brown JA, et al. Enhancement of Zika virus pathogenesis by preexisting antinflavivirus immunity. *Science* 2017;356:175–80.
- [48] Smalley C, Erasmus JH, Chesson CB, Beasley DWC. In: Status of research and development of vaccines for chikungunya Vaccine, vol. 34; 2016. p. 2976–81.
- [49] Vannice KS, Durbin A, Hombach J Status of vaccine research and development of vaccines for dengue Vaccine 2016;34:2934–8.
- [50] Vaccine Dengue. WHO position paper. Weekly epidemiological position paper 2018;93:457–76. https://www.who.int/immunization/policy/position_papers/dengue/en/.
- [51] World Health Organization. https://www.who.int/immunization/research/development/WHO_UNICEF_Zikavac_TPP_Feb2017.pdf?ua=1.
- [52] Barrett ADT. Current status of Zika vaccine development. Zika vaccines advance into clinical evaluation. *NPJ Vaccines* 2018;3:24. <https://doi.org/10.1038/s41541-018-0061-9>.
- [53] Watanabe S, Wei Wen Tan N, Wing Ki Chan K, Vasudevan SG. Dengue Virus and Zika Virus Serological Cross-reactivity and Their Impact on Pathogenesis in Mice. *J Infect Dis* 2019;219:223–33. <https://doi.org/10.1093/infdis/jiy482>.