Published in final edited form as:

Curr Opin Immunol. 2018 August ; 53: 130–136. doi:10.1016/j.coi.2018.04.024.

Zika Virus Vaccines: Immune Response, Current Status, and Future Challenges

Justin M. Richner¹ and Michael S. Diamond^{2,3,4,5,*}

¹Department of Microbiology & Molecular Biology, IIT Research Institute, Chicago, IL 60616, USA

²Department of Medicine, Washington University School of Medicine, St. Louis, MO 63110, USA

³Department of Pathology and Immunology, Washington University School of Medicine, St. Louis, MO 63110, USA

⁴Department of Molecular Microbiology, Washington University School of Medicine, St. Louis, MO 63110, USA

⁵The Andrew M. and Jane M. Bursky Center for Human Immunology and Immunotherapy Programs, Washington University School of Medicine, St. Louis, MO 63110, USA

Abstract

Zika virus (ZIKV) is the most recent mosquito-transmitted virus to cause a global health crisis following its entrance into a naïve population in the Western Hemisphere. Once the ZIKV outbreak began investigators rapidly established small and large animal models of pathogenesis, developed a number candidate vaccines using different platforms, and defined mechanisms of protection. In this review, we characterize the adaptive immune response elicited by ZIKV infections and vaccines, the status of ongoing clinical trials in humans, and discuss future challenges within the field.

Introduction

Zika virus (ZIKV) is an enveloped, positive-sense single stranded RNA virus within the Flavivirus genus of the *Flaviviridae*. This family encompasses a number of globally relevant viruses that have been targeted for vaccine development including yellow fever (YFV) [1], Japanese encephalitis (JEV) [2], Dengue (DENV) [3], West Nile (WNV) [4], and tick-borne encephalitis (TBEV) viruses [5]. The RNA genome of these viruses encodes for a single open reading frame, which is translated as a polyprotein and cleaved by viral and cellular proteases to generate the three structural proteins (capsid (C), pre-membrane (prM), and envelope (E)) and seven non-structural proteins (NS1, NS2A, NS2B, NS3, NS4A, NS4B,

*Address correspondence to: Michael S. Diamond, M.D. Ph.D., diamond@wusm.wustl.edu.

POTENTIAL CONFLICTS OF INTEREST

MSD is a consultant for Inbios and Sanofi-Pasteur, and is on the Scientific Advisory Board of Moderna.

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

and NS5). Neutralizing antibodies elicited upon infection or vaccination predominantly target the E protein on the surface of the viral particle [6, 7]. Prior to 2007, ZIKV was a relatively obscure virus and associated with mild febrile illness in a small number of human cases. In 2007, the virus spread to the Pacific Islands where it caused large outbreaks. ZIKV entered northeastern Brazil in 2013 [8] and spread over the following two years throughout Central and South America, and into parts of North America, including the United States. From 2015 to January 2018, more than 800,000 documented and suspected human cases of ZIKV were reported [9]. In addition to an explosion of new human cases, the outbreak was associated with congenital defects in the context of infection of pregnant women, which resulted in publicized cases of microcephaly [10] and other birth defects [11]. In adults, ZIKV also has been linked to an increased incidence of Guillain-Barré syndrome [12–14], an autoimmune, neurological disorder characterized by ascending muscle weakness. The emergence of ZIKV into a new geographical niche along with novel viral-associated diseases prompted the call for medical countermeasures. In response, several vaccine platforms have been developed rapidly including mRNA, DNA, virus-vectored, live-attenuated viruses, and inactivated viral particles, and are described in recent reviews [15–17]. Here, we discuss the adaptive immune responses to vaccination in the context of animal models, the preliminary data from human clinical trials (Table 1), and the possible challenges facing the field (Figure 1).

Adaptive immune responses to ZIKV infection and vaccination

The success of a viral vaccine depends on its ability to induce durable adaptive B and T cell responses that inhibit viral replication and dissemination. The observation that a primary ZIKV infection in rhesus macaques inhibited secondary challenge with a homologous or heterologous virus [18–20] suggests that anti-ZIKV adaptive immunity can be protective. Many of the current suite of anti-ZIKV vaccines induce high levels of neutralizing antibody and antiviral CD4⁺ and CD8⁺ T cell responses. Antibody alone is sufficient for protection, as passive transfer of IgG from purified formalin-inactivated virus (PIV) vaccinated non-human primates (NHPs) into naïve recipient mice or NHPs inhibited ZIKV viremia upon challenge [21]. Similarly, passive transfer of sera isolated from humans vaccinated with PIV or a DNA vaccine encoding the prM-E viral genes blocked viremia [22] and protected immunodeficient mice [23]. Defining the threshold neutralization titer required for vaccine immunity is critical for determining a correlate of protection. A threshold neutralizing titer in serum of approximately 1/100 [21, 24, 25] to 1/1000 [26] inhibited viremia following viral challenge after vaccination of NHPs. In human clinical trials, three doses of a DNA vaccine encoding for prM-E, or two doses of the PIV vaccine achieved mean titers above this 1/100 threshold [22, 27]. Notwithstanding these results, the level of neutralizing antibody required for sterilizing immunity is much higher. In immunodeficient mice, the neutralizing antibody titer that prevented virus detection in serum or susceptible tissues and an anamnestic immune response was greater than 1/10,000 [28]. A key question remains as to whether sterilizing immunity is required to prevent seeding of the placenta and vertical transmission.

Characterization of the circulating plasmablasts and memory B cells from ZIKV-infected human patients has helped to define epitope targets on the E protein of strongly neutralizing

and protective mAbs [29–33]. Antibodies elicited against the lateral ridge epitope in domain III, quaternary epitopes spanning domains I, II, and III across the E protein dimer (EDE mAbs), and sites within domains I or II were highly neutralizing [30–32, 34, 35]. In contrast, other highly cross-reactive antibodies that targeted the conserved fusion loop in domain II were poorly neutralizing and could lead to antibody-dependent enhancement of infection (ADE, see below) [29, 36]. Likely, different vaccine platforms induce antibodies and memory B cells with unique epitope specificity, which may have implications for protective activity and cross-reactivity. A structural correlate of antibody protection is an important goal where mapping epitopes of vaccine-induced polyclonal antibodies can be related to long-term efficacy.

In addition to B-cell mediated immunity, T cells also contribute to protection against ZIKV infection. Polyfunctional antiviral CD4⁺ and CD8⁺ T cells can be detected after ZIKV infection in mice, NHPs, and humans [19, 37–40], with many of the immunodominant epitopes within the structural genes (E, prM, and C) [37]. In susceptible mouse models, the absence of CD8⁺ T cells led to increased viral burden and mortality, and adoptive transfer of T cells from infected mice limited viral infection in a CD8⁺ T cell deficient host [38, 39]. Vaccines also induce antiviral T cell responses. Vaccination of mice with DNA or mRNA vaccines encoding for prM-E or vaccination of NHPs with PIV, DNA, or adenovirus-vectored vaccines induced antiviral CD4⁺ and CD8⁺ T cells [21, 25, 40, 41]. However, depletion of CD4⁺ and/or CD8⁺ T cells in mice that were immunized with a DNA vaccine had no effect on protective immunity [41], suggesting that while T cells contribute to the adaptive immune response, they may not be absolutely necessary for protection. In preliminary data from human clinical trials with DNA vaccines, anti-E CD4⁺ and CD8⁺ cytokine-secreting cells were detected following peptide stimulation [27].

The impact of antibody dependent enhancement on ZIKV vaccines

Antibodies that bind to but fail to neutralize the closely related DENV can lead to increased infection in FcγR-expressing cells and higher systemic viral titers through a phenomenon known as ADE. DENV disease severity has been linked to ADE in the context of secondary infections with heterologous DENV serotypes [42, 43]. The contribution of pre-existing flavivirus immunity and ADE on ZIKV pathogenesis has been debated. Although cross-reactive antibodies against the fusion loop in the E protein that were generated in the context of heterologous flavivirus infection or vaccination can enhance ZIKV infection in cell culture and mice [29, 44–46], this phenomenon has not yet been observed in NHPs. Challenge of DENV-immune rhesus macaques with ZIKV did not result in enhanced viral burden or disease [47, 48]. Furthermore, preliminary epidemiological evidence in humans suggests no enhancement of ZIKV disease in those with pre-existing DENV immunity compared to naïve individuals [49]. The contribution of DENV-immunity towards vaccine efficacy has not been explored, but prior DENV-1 exposure has been linked to greater neutralizing titers upon a secondary ZIKV challenge in humans [31].

The effect of the reciprocal sequence, pre-existing ZIKV immunity on DENV pathogenesis, also remains uncertain, yet clearly has implications for ZIKV vaccine campaigns in DENV endemic areas. Monoclonal or polyclonal antibodies derived from ZIKV infection or

vaccination can enhance DENV infection in cell culture and pathogenesis *in vivo* in some mouse strains [28, 29]. More recent studies in NHPs have demonstrated that prior ZIKV infection can lead to enhanced viral titers and elevated cytokine levels following DENV-2 challenge [50]. Many of the current vaccine platforms under development encode for the envelope genes and are capable of eliciting cross-reactive antibodies targeting the fusion loop in domain II [22, 23, 27]. Given this approach, the possible enhancement of DENV infection by prior ZIKV immunization may need to be considered during vaccine development. In future NHP studies and clinical trials, careful evaluation and titration of prior flavivirus antibody levels will be critical for defining the contribution of antibody cross-reactivity towards vaccine efficacy and enhanced pathogenesis through ADE. Related to this idea, immunization with the tetravalent DENV vaccine Dengvaxia® was halted in the Philippines, after it was discovered that naïve children receiving the vaccine were more susceptible to severe Dengue complications compared to unvaccinated children [51]. Researchers and manufacturers need to ensure that the ZIKV vaccines are safe for naïve populations in endemic areas.

In the interim, several groups have developed strategies to limit the flavivirus cross-reactivity in the context of a ZIKV vaccine. One study showed that elimination of the conserved fusion loop epitope in a prM-E vaccine reduced ADE of DENV *in vitro* and *in vivo* despite inducing comparable levels of neutralizing antibodies against ZIKV [28]. Other vaccines have targeted regions of the viral proteome that do not elicit ADE [52, 53]. A vaccinia-vectored vaccine encoding for the non-structural protein NS1 induced protective CD8⁺ T cells and anti-NS1 antibody responses [52]. Because NS1 is not expressed on the surface of the viral particle, elicited antibodies do not promote enhanced viral uptake by the FcγR-bearing cells.

Other considerations of ZIKV vaccines

The mechanism leading to the increased incidence of Guillain-Barré syndrome (GBS) associated with ZIKV infections is unknown, although several hypotheses have been proposed [54]. In an *ex vivo* culture system, ZIKV infected myelinating oligodendrocytes leading to reduced myelin staining, suggesting a direct involvement of virus infection on disrupted myelin, which could contribute to GBS [55]. With this mechanism, vaccines limiting ZIKV infection should lower the risk of GBS. Alternatively, as ZIKV structural and non-structural proteins share possible epitopes with proteins associated with myelination and axonal neuropathy [56], the induction of cross-reactive antibodies could lead to GBS. In this case, ZIKV vaccines containing these conserved epitopes could increase the risk of GBS. Further research defining the underlying mechanisms of GBS is needed to help predict the safety and outcome of ZIKV vaccination.

The greatest threat to human health posed by ZIKV is damage to the fetus during pregnancy. To date, two vaccines have been tested in pregnancy, and these studies were performed exclusively in mice: mRNA vaccines encoding for the prM-E genes and live-attenuated ZIKV strains largely prevented virus dissemination into the maternal decidua and fetal placenta, which prevented virus infection of the fetal brain [57, 58]. Other studies have demonstrated that neonatal mice born to vaccinated mothers were protected from a lethal

ZIKV challenge [59]. Based on these studies, it is likely that vaccines inducing high levels of neutralizing antibody can reduce or block vertical transmission. Nonetheless, the titer of neutralizing antibody required to prevent ZIKV spread to the developing fetus remains to be determined for NHPs and humans. Even high levels of serum neutralizing antibody following vaccination or passive transfer of monoclonal antibodies did not completely protect all pregnant mice from vertical transmission, as some placenta and fetal heads still had low levels of ZIKV RNA [30, 36, 57, 58]. Virus-infected myeloid cells in blood [60] that are not neutralized by antibody might seed the placenta, which would make complete inhibition of vertical transmission by serum antibodies challenging. The level of virus in the fetal brain that is necessary to cause developmental abnormalities has not been determined, but it may be low, as babies born to NHP mothers asymptomatically infected with ZIKV still displayed developmental defects and loss of neuronal stem cells [61]. Thus, the magnitude and constituent vaccine immune response necessary to block vertical transmission requires further evaluation.

Current trials and future perspectives

There are 13 ongoing clinical trials (Table 1) of ZIKV vaccines utilizing multiple platforms. All of these strategies show efficacy in murine models, and many block infections in NHPs. Live-attenuated vaccines and adenoviral vectors generate the most robust immune responses following a single administration [21, 24, 41, 62], whereas DNA, mRNA, and purified inactivated vaccines elicit robust immunity following a prime-boost schedule [21, 24, 28, 41]. In NHPs, immunity following DNA vaccination waned over time whereas responses of purified inactivated virus or adenovirus-vectored vaccines remained protective through one year post immunization [24]. Preliminary data from clinical trials have been published on different prM-E DNA vaccines [23, 27] and a PIV vaccine [22]. All trials showed good safety profiles with only mild reactogenicity, and achieved >90% seroconversion rates with participants receiving the optimal dosing regimens. Both DNA platforms induced antiviral T cell responses by intracellular cytokine staining [27] or ELISPOT analysis [23]. A direct comparison of neutralization titers across studies is difficult as different methodology was used. A concerted effort to normalize neutralization methods may be needed to define universal correlates of protection for upcoming efficacy trials. A ZIKV vaccine likely will be targeted to areas with endemic Dengue virus; however the contribution of pre-existing flavivirus immunity to vaccine efficacy has not been evaluated. Retrospective analysis of individuals in the PIV trial revealed only a few participants had pre-existing immunity against other flaviviruses [22]. Although the sample size was small, these individuals had increased antibody titers relative to flavivirus-naïve individuals suggesting that pre-existing DENV immunity might enhance vaccine efficacy. Ongoing trials NCT02887482, NCT03110770, NCT02963909, NCT03008122, and NCT03343626 are designed to address this question.

Conclusion

In under three years, it is remarkable that many different ZIKV vaccine platforms have advanced to human clinical testing. A significant challenge facing the licensure of a ZIKV vaccine may be demonstrating vaccine efficacy in a phase III clinical trials with declining

rates of ZIKV transmission. Efficacy studies will need to be designed for rapid enrollment in areas with outbreaks of ZIKV transmission. Additionally, data in NHPs likely will be needed to support clinical trial data. Demonstration that ZIKV vaccines can prevent vertical transmission and congenital Zika syndrome may be challenging if outbreaks remain small and dispersed. Beyond this, the introduction of a ZIKV vaccine to combat natural infection and congenital disease will need to balance efficacy with the theoretical risks of immune enhancement of DENV or GBS.

Acknowledgments

FUNDING

This work was supported by grants from the NIH (R01 AI073755, R01 AI127828, and R01 HD091218).

References

1. Beck AS, Barrett AD. Current status and future prospects of yellow fever vaccines. *Expert Rev Vaccines*. 2015; 14(11):1479–92. DOI: 10.1586/14760584.2015.1083430 [PubMed: 26366673]
2. Yun SI, Lee YM. Japanese encephalitis: the virus and vaccines. *Hum Vaccin Immunother*. 2014; 10(2):263–79. DOI: 10.4161/hv.26902 [PubMed: 24161909]
3. Katzelnick LC, Coloma J, Harris E. Dengue: knowledge gaps, unmet needs, and research priorities. *Lancet Infect Dis*. 2017; 17(3):e88–e100. DOI: 10.1016/S1473-3099(16)30473-X [PubMed: 28185868]
4. Amanna IJ, Slifka MK. Current trends in West Nile virus vaccine development. *Expert Rev Vaccines*. 2014; 13(5):589–608. DOI: 10.1586/14760584.2014.906309 [PubMed: 24689659]
5. Kollaritsch H, Paulke-Korinek M, Holzmann H, Hombach J, Bjorvatn B, Barrett A. Vaccines and vaccination against tick-borne encephalitis. *Expert Rev Vaccines*. 2012; 11(9):1103–19. DOI: 10.1586/erv.12.86 [PubMed: 23151167]
6. Sirohi D, Chen Z, Sun L, Klose T, Pierson TC, Rossmann MG, et al. The 3.8 Å resolution cryo-EM structure of Zika virus. *Science*. 2016; 352(6284):467–70. DOI: 10.1126/science.aaf5316 [PubMed: 27033547]
7. Weaver SC, Costa F, Garcia-Blanco MA, Ko AI, Ribeiro GS, Saade G, et al. Zika virus: History, emergence, biology, and prospects for control. *Antiviral Res*. 2016; 130:69–80. DOI: 10.1016/j.antiviral.2016.03.010 [PubMed: 26996139]
8. Grubaugh ND, Faria NR, Andersen KG, Pybus OG. Genomic Insights into Zika Virus Emergence and Spread. *Cell*. 2018; 172(6):1160–2. DOI: 10.1016/j.cell.2018.02.027 [PubMed: 29522736]
9. Zika cases and congenital syndrome associated with Zika virus reported by countries and territories in the Americas, 2015–2018 Cumulative cases [Internet]. Pan American Health Organization - World Health Organization; [cited March 16, 2018]. Available from: http://www.paho.org/hq/index.php?option=com_content&view=article&id=12390&Itemid=42090&lang=en
10. Mlakar J, Korva M, Tul N, Popovic M, Poljsak-Prijatelj M, Mraz J, et al. Zika Virus Associated with Microcephaly. *N Engl J Med*. 2016; 374(10):951–8. DOI: 10.1056/NEJMoa1600651 [PubMed: 26862926]
11. Coyne CB, Lazear HM. Zika virus - reigniting the TORCH. *Nature reviews Microbiology*. 2016; 14(11):707–15. DOI: 10.1038/nrmicro.2016.125 [PubMed: 27573577]
12. Parra B, Lizarazo J, Jimenez-Arango JA, Zea-Vera AF, Gonzalez-Manrique G, Vargas J, et al. Guillain-Barre Syndrome Associated with Zika Virus Infection in Colombia. *N Engl J Med*. 2016; 375(16):1513–23. DOI: 10.1056/NEJMoa1605564 [PubMed: 27705091]
13. Krauer F, Riesen M, Reveiz L, Oladapo OT, Martinez-Vega R, Porgo TV, et al. Zika Virus Infection as a Cause of Congenital Brain Abnormalities and Guillain-Barre Syndrome: Systematic Review. *PLoS Med*. 2017; 14(1):e1002203.doi: 10.1371/journal.pmed.1002203 [PubMed: 28045901]

14. Cao-Lormeau VM, Blake A, Mons S, Lastere S, Roche C, Vanhomwegen J, et al. Guillain-Barre Syndrome outbreak associated with Zika virus infection in French Polynesia: a case-control study. *Lancet*. 2016; 387(10027):1531–9. DOI: 10.1016/S0140-6736(16)00562-6 [PubMed: 26948433]
15. Makhluaf H, Shresta S. Development of Zika Virus Vaccines. *Vaccines (Basel)*. 2018; 6(1)doi: 10.3390/vaccines6010007
16. Fernandez E, Diamond MS. Vaccination strategies against Zika virus. *Curr Opin Virol*. 2017; 23:59–67. DOI: 10.1016/j.coviro.2017.03.006 [PubMed: 28432975]
17. McArthur MA. Zika Virus: Recent Advances towards the Development of Vaccines and Therapeutics. *Viruses*. 2017; 9(6)doi: 10.3390/v9060143
18. Aliota MT, Dudley DM, Newman CM, Mohr EL, Gellerup DD, Breitbach ME, et al. Heterologous Protection against Asian Zika Virus Challenge in Rhesus Macaques. *PLoS Negl Trop Dis*. 2016; 10(12):e0005168.doi: 10.1371/journal.pntd.0005168 [PubMed: 27911897]
19. Dudley DM, Aliota MT, Mohr EL, Weiler AM, Lehrer-Brey G, Weisgrau KL, et al. A rhesus macaque model of Asian-lineage Zika virus infection. *Nat Commun*. 2016; 7:12204.doi: 10.1038/ncomms12204 [PubMed: 27352279]
20. Osuna CE, Lim SY, Deleage C, Griffin BD, Stein D, Schroeder LT, et al. Zika viral dynamics and shedding in rhesus and cynomolgus macaques. *Nature medicine*. 2016; 22(12):1448–55. DOI: 10.1038/nm.4206
21. Abbink P, Larocca RA, De La Barrera RA, Bricault CA, Moseley ET, Boyd M, et al. Protective efficacy of multiple vaccine platforms against Zika virus challenge in rhesus monkeys. *Science*. 2016; 353(6304):1129–32. DOI: 10.1126/science.aah6157 [PubMed: 27492477]
- 22. Modjarrad K, Lin L, George SL, Stephenson KE, Eckels KH, De La Barrera RA, et al. Preliminary aggregate safety and immunogenicity results from three trials of a purified inactivated Zika virus vaccine candidate: phase 1, randomised, double-blind, placebo-controlled clinical trials. *Lancet*. 2017; A PIV vaccine is safe and immunogenic in patients within ongoing clinical trials. Peak neutralization titers reached over 1/100. Patients with pre-existing flavivirus immunity had elevated neutralizing titers compared to flavivirus-naïve patients. doi: 10.1016/S0140-6736(17)33106-9
- 23. Tebas P, Roberts CC, Muthumani K, Reuschel EL, Kudchodkar SB, Zaidi FI, et al. Safety and Immunogenicity of an Anti-Zika Virus DNA Vaccine - Preliminary Report. *N Engl J Med*. 2017; A DNA vaccine encoding for prM-E is safe and immunogenic in patients within an ongoing clinical trial. Three doses of a one or two miligram immunization yielded anti-E antibody titers of 1/2000 and 100% seroconversion as well as antiviral T cells. doi: 10.1056/NEJMoa1708120
- 24. Abbink P, Larocca RA, Visitsunthorn K, Boyd M, De La Barrera RA, Gromowski GD, et al. Durability and correlates of vaccine protection against Zika virus in rhesus monkeys. *Science translational medicine*. 2017; 9(420) The authors vaccinated mice and NHPs with adenovirus-vectored, DNA, and PIV vaccines. PIV and adenovirus-vectored vaccines yielded similar immune responses that were durable and protective over one year. Humoral responses post DNA vaccination waned to below protective levels. doi: 10.1126/scitranslmed.aao4163
25. Pardi N, Hogan MJ, Pelc RS, Muramatsu H, Andersen H, DeMaso CR, et al. Zika virus protection by a single low-dose nucleoside-modified mRNA vaccination. *Nature*. 2017; 543(7644):248–51. DOI: 10.1038/nature21428 [PubMed: 28151488]
26. Dowd KA, Ko SY, Morabito KM, Yang ES, Pelc RS, DeMaso CR, et al. Rapid development of a DNA vaccine for Zika virus. *Science*. 2016; 354(6309):237–40. DOI: 10.1126/science.aai9137 [PubMed: 27708058]
- 27. Gaudinski MR, Houser KV, Morabito KM, Hu Z, Yamshchikov G, Rothwell RS, et al. Safety, tolerability, and immunogenicity of two Zika virus DNA vaccine candidates in healthy adults: randomised, open-label, phase 1 clinical trials. *Lancet*. 2017; DNA vaccines encoding for variants of prM-E with different doses are safe and immunogenic in patients within ongoing clinical trials. Patients immunized with three doses across two injection sites of a DNA vaccine encoding for ZIKV prM-E had superior immune responses with mean neutralizing titers of 1/300 and antiviral CD4⁺ and CD8⁺ T cells detected by intracellular cytokine staining. doi: 10.1016/S0140-6736(17)33105-7
- 28. Richner JM, Himansu S, Dowd KA, Butler SL, Salazar V, Fox JM, et al. Modified mRNA Vaccines Protect against Zika Virus Infection. *Cell*. 2017; 168(6):1114–25. e10. An mRNA

- vaccine encoding for ZIKV prM-E yielded robust humoral immunity and protection in murine models. Immunization with the wild-type prM-E led to *in vitro* and *in vivo* ADE, whereas ADE was diminished greatly following immunization with a prM-E mutant lacking the conserved fusion loop epitope of the E protein. DOI: 10.1016/j.cell.2017.02.017 [PubMed: 28222903]
29. Stettler K, Beltramello M, Espinosa DA, Graham V, Cassotta A, Bianchi S, et al. Specificity, cross-reactivity, and function of antibodies elicited by Zika virus infection. *Science*. 2016; 353(6301): 823–6. DOI: 10.1126/science.aaf8505 [PubMed: 27417494]
 30. Sapparapu G, Fernandez E, Kose N, Bin C, Fox JM, Bombardi RG, et al. Neutralizing human antibodies prevent Zika virus replication and fetal disease in mice. *Nature*. 2016; 540(7633):443–7. DOI: 10.1038/nature20564 [PubMed: 27819683]
 31. Robbiani DF, Bozzacco L, Keeffe JR, Khouri R, Olsen PC, Gazumyan A, et al. Recurrent Potent Human Neutralizing Antibodies to Zika Virus in Brazil and Mexico. *Cell*. 2017; 169(4):597–609. e11. DOI: 10.1016/j.cell.2017.04.024 [PubMed: 28475892]
 32. Wang Q, Yang H, Liu X, Dai L, Ma T, Qi J, et al. Molecular determinants of human neutralizing antibodies isolated from a patient infected with Zika virus. *Science translational medicine*. 2016; 8(369):369ra179.doi: 10.1126/scitranslmed.aai8336
 33. Rogers TF, Goodwin EC, Briney B, Sok D, Beutler N, Strubel A, et al. Zika virus activates de novo and cross-reactive memory B cell responses in dengue-experienced donors. *Sci Immunol*. 2017; 2(14)doi: 10.1126/sciimmunol.aan6809
 34. Dai L, Song J, Lu X, Deng YQ, Musyoki AM, Cheng H, et al. Structures of the Zika Virus Envelope Protein and Its Complex with a Flavivirus Broadly Protective Antibody. *Cell Host Microbe*. 2016; 19(5):696–704. DOI: 10.1016/j.chom.2016.04.013 [PubMed: 27158114]
 35. Swanstrom JA, Plante JA, Plante KS, Young EF, McGowan E, Gallichotte EN, et al. Dengue Virus Envelope Dimer Epitope Monoclonal Antibodies Isolated from Dengue Patients Are Protective against Zika Virus. *MBio*. 2016; 7(4)doi: 10.1128/mBio.01123-16
 36. Zhao H, Fernandez E, Dowd KA, Speer SD, Platt DJ, Gorman MJ, et al. Structural Basis of Zika Virus-Specific Antibody Protection. *Cell*. 2016; 166(4):1016–27. DOI: 10.1016/j.cell.2016.07.020 [PubMed: 27475895]
 37. Grifoni A, Pham J, Sidney J, O'Rourke PH, Paul S, Peters B, et al. Prior Dengue virus exposure shapes T cell immunity to Zika virus in humans. *Journal of virology*. 2017; doi: 10.1128/JVI.01469-17
 38. Huang H, Li S, Zhang Y, Han X, Jia B, Liu H, et al. CD8(+) T Cell Immune Response in Immunocompetent Mice during Zika Virus Infection. *Journal of virology*. 2017; 91(22)doi: 10.1128/JVI.00900-17
 39. Elong Ngono A, Vizcarra EA, Tang WW, Sheets N, Joo Y, Kim K, et al. Mapping and Role of the CD8(+) T Cell Response During Primary Zika Virus Infection in Mice. *Cell Host Microbe*. 2017; 21(1):35–46. DOI: 10.1016/j.chom.2016.12.010 [PubMed: 28081442]
 40. Muthumani K, Griffin BD, Agarwal S, Kudchodkar SB, Reuschel EL, Choi H, et al. In vivo protection against ZIKV infection and pathogenesis through passive antibody transfer and active immunisation with a prMEnv DNA vaccine. 2016; 1:16021.doi: 10.1038/npjvaccines.2016.21
 41. Larocca RA, Abbink P, Peron JP, Zannotto PM, Iampietro MJ, Badamchi-Zadeh A, et al. Vaccine protection against Zika virus from Brazil. *Nature*. 2016; 536(7617):474–8. DOI: 10.1038/nature18952 [PubMed: 27355570]
 42. Katzelnick LC, Gresh L, Halloran ME, Mercado JC, Kuan G, Gordon A, et al. Antibody-dependent enhancement of severe dengue disease in humans. *Science*. 2017; 358(6365):929–32. DOI: 10.1126/science.aan6836 [PubMed: 29097492]
 43. Halstead SB. Neutralization and antibody-dependent enhancement of dengue viruses. *Adv Virus Res*. 2003; 60:421–67. [PubMed: 14689700]
 44. Bardina SV, Bunduc P, Tripathi S, Duehr J, Frere JJ, Brown JA, et al. Enhancement of Zika virus pathogenesis by preexisting anti-flavivirus immunity. *Science*. 2017; 356(6334):175–80. DOI: 10.1126/science.aal4365 [PubMed: 28360135]
 45. Priyamvada L, Quicke KM, Hudson WH, Onlamoon N, Sewatanon J, Edupuganti S, et al. Human antibody responses after dengue virus infection are highly cross-reactive to Zika virus.

- Proceedings of the National Academy of Sciences of the United States of America. 2016; 113(28): 7852–7. DOI: 10.1073/pnas.1607931113 [PubMed: 27354515]
46. Dejnirattisai W, Supasa P, Wongwiwat W, Rouvinski A, Barba-Spaeth G, Duangchinda T, et al. Dengue virus sero-cross-reactivity drives antibody-dependent enhancement of infection with Zika virus. *Nature immunology*. 2016; 17(9):1102–8. DOI: 10.1038/ni.3515 [PubMed: 27339099]
 47. Pantoja P, Perez-Guzman EX, Rodriguez IV, White LJ, Gonzalez O, Serrano C, et al. Zika virus pathogenesis in rhesus macaques is unaffected by pre-existing immunity to dengue virus. *Nat Commun*. 2017; 8:15674.doi: 10.1038/ncomms15674 [PubMed: 28643775]
 48. McCracken MK, Gromowski GD, Friberg HL, Lin X, Abbink P, De La Barrera R, et al. Impact of prior flavivirus immunity on Zika virus infection in rhesus macaques. *PLoS pathogens*. 2017; 13(8):e1006487.doi: 10.1371/journal.ppat.1006487 [PubMed: 28771605]
 49. Terzian ACB, Schanoski AS, Mota MTO, da Silva RA, Estofolete CF, Colombo TE, et al. Viral Load and Cytokine Response Profile Does Not Support Antibody-Dependent Enhancement in Dengue-Primed Zika Virus-Infected Patients. *Clin Infect Dis*. 2017; 65(8):1260–5. DOI: 10.1093/cid/cix558 [PubMed: 29017246]
 50. George J, Valiant WG, Mattapallil MJ, Walker M, Huang YS, Vanlandingham DL, et al. Prior Exposure to Zika Virus Significantly Enhances Peak Dengue-2 Viremia in Rhesus Macaques. *Sci Rep*. 2017; 7(1):10498.doi: 10.1038/s41598-017-10901-1 [PubMed: 28874759]
 51. Halstead SB. Safety issues from a Phase 3 clinical trial of a live-attenuated chimeric yellow fever tetravalent dengue vaccine. *Hum Vaccin Immunother*. 2018; doi: 10.1080/21645515.2018.1445448
 52. Brault AC, Domi A, McDonald EM, Talmi-Frank D, McCurley N, Basu R, et al. A Zika Vaccine Targeting NS1 Protein Protects Immunocompetent Adult Mice in a Lethal Challenge Model. *Sci Rep*. 2017; 7(1):14769.doi: 10.1038/s41598-017-15039-8 [PubMed: 29116169]
 53. Yang M, Dent M, Lai H, Sun H, Chen Q. Immunization of Zika virus envelope protein domain III induces specific and neutralizing immune responses against Zika virus. *Vaccine*. 2017; 35(33): 4287–94. DOI: 10.1016/j.vaccine.2017.04.052 [PubMed: 28669618]
 54. Rodriguez Y, Rojas M, Pacheco Y, Acosta-Ampudia Y, Ramirez-Santana C, Monsalve DM, et al. Guillain-Barre syndrome, transverse myelitis and infectious diseases. *Cellular & molecular immunology*. 2018; doi: 10.1038/cmi.2017.142
 55. Cumberworth SL, Barrie JA, Cunningham ME, de Figueiredo DPG, Schultz V, Wilder-Smith AJ, et al. Zika virus tropism and interactions in myelinating neural cell cultures: CNS cells and myelin are preferentially affected. *Acta Neuropathol Commun*. 2017; 5(1):50.doi: 10.1186/s40478-017-0450-8 [PubMed: 28645311]
 56. Lucchese G, Kanduc D. Zika virus and autoimmunity: From microcephaly to Guillain-Barre syndrome, and beyond. *Autoimmun Rev*. 2016; 15(8):801–8. DOI: 10.1016/j.autrev.2016.03.020 [PubMed: 27019049]
 57. Richner JM, Jagger BW, Shan C, Fontes CR, Dowd KA, Cao B, et al. Vaccine Mediated Protection Against Zika Virus-Induced Congenital Disease. *Cell*. 2017; 170(2):273–83.e12. DOI: 10.1016/j.cell.2017.06.040 [PubMed: 28708997]
 58. Shan C, Muruato AE, Jagger BW, Richner J, Nunes BTD, Medeiros DBA, et al. A single-dose live-attenuated vaccine prevents Zika virus pregnancy transmission and testis damage. *Nat Commun*. 2017; 8(1):676.doi: 10.1038/s41467-017-00737-8 [PubMed: 28939807]
 59. Kim E, Erdos G, Huang S, Kenniston T, Falo LD Jr, Gambotto A. Preventative Vaccines for Zika Virus Outbreak: Preliminary Evaluation. *EBioMedicine*. 2016; 13:315–20. DOI: 10.1016/j.ebiom.2016.09.028 [PubMed: 27717627]
 60. Michlmayr D, Andrade P, Gonzalez K, Balmaseda A, Harris E. CD14(+)CD16(+) monocytes are the main target of Zika virus infection in peripheral blood mononuclear cells in a paediatric study in Nicaragua. *Nat Microbiol*. 2017; 2(11):1462–70. DOI: 10.1038/s41564-017-0035-0 [PubMed: 28970482]
 61. Adams Waldorf KM, Nelson BR, Stencel-Baerenwald JE, Studholme C, Kapur RP, Armistead B, et al. Congenital Zika virus infection as a silent pathology with loss of neurogenic output in the fetal brain. *Nature medicine*. 2018; 24(3):368–74. DOI: 10.1038/nm.4485

62. Shan C, Muruato AE, Nunes BTD, Luo H, Xie X, Medeiros DBA, et al. A live-attenuated Zika virus vaccine candidate induces sterilizing immunity in mouse models. *Nature medicine*. 2017; 23(6):763–7. DOI: 10.1038/nm.4322
63. Zika - Epidemiological Update 25 August 2017 [Internet]. Pan American Health Organization - World Health Organization;

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

HIGHLIGHTS

- B and T cell-mediated adaptive immune responses inhibit ZIKV in animal models
- Early phase clinical trials in humans have demonstrated safety and immunogenicity
- ZIKV vaccines may need to consider potential enhancement of DENV infection and GBS

Figure 1. Challenges and questions facing the Zika vaccine field

A. The infection rate has declined since the peak in 2016, which affects vaccine efficacy study design. This graph shows the number of new ZIKV cases each week (adapted from [63]). **B.** Pre-existing immunity against DENV or other flaviviruses could inhibit or skew the immune response against ZIKV vaccines. **C.** ZIKV infection has been linked to Guillain-Barré Syndrome. In theory, vaccination might induce antiviral antibodies that cross-react with host proteins on glial cells and lead to autoimmunity and GBS. **D.** ZIKV vaccines share conserved epitopes with related flaviviruses, including DENV. Vaccine-induced anti-ZIKV antibodies that cross-react with DENV (e.g., fusion loop in domain II) could facilitate infection in FcγR-expressing cells leading to ADE and worsened DENV disease. In this Figure, antibodies of different colors represent distinct clones of a polyclonal humoral response.

Table 1

ZIKV vaccines in clinical trials

Vaccine	Platform	Sponsors	Clinical Trials	Status	References
mRNA-1325	mRNA encoding prM-E	Moderna Therapeutics BARDA ^a	NCT03014089	Phase I/II, Ongoing	Richner et al [28] Richner et al [57]
GLS-5700	DNA encoding prM-E	GeneOne Life Sciences Inovio Pharmaceuticals	NCT02809443 NCT02887482	Phase I Ongoing Phase I Ongoing	Tebas et al [23] Muthumani et al [40]
VRC5288	DNA encoding prM-E (Chimeric ZIKV-JEV)	NIAID ^b	NCT02840487	Phase I Ongoing	Gaudinski et al [27] Dowd et al [26]
VRC5283	DNA encoding prM-E (ZIKV)	NIAID	NCT02996461 NCT03110770	Phase I, Ongoing Phase II, Enrolling	Gaudinski et al [27] Dowd et al [26]
ZPIV	Purified inactivated virus	NIAID WRAIR ^c	NCT02963909 NCT02952833 NCT02937233 NCT03008122	Phase I, Ongoing Phase I, Ongoing Phase I, Ongoing Phase I, Enrolling	Modjarrad et al [22] Larocca et al [41] Abbink et al [21]
VLA1601	Purified inactivated virus	ValNeva Emergent BioSolutions	NCT03425149	Phase I, Enrolling	
TAK-426	Purified inactivated virus	Takeda BARDA	NCT03343626	Phase I, Enrolling	
MV-ZIKA	Measles-vectored	Themis Biosciences	NCT02996890	Phase I, Ongoing	

^aBARDA = Biomedical Advanced Research & Development Authority

^bNIAID = National Institute of Allergy & Infectious Diseases

^cWRAIR = Walter Reed Army Institute of Research