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REVIEW

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Profile of clinical research related to the development of the Chikungunya virus vaccine: strategic points and challenges

Perfil das pesquisas clínicas relacionadas ao desenvolvimento da vacina contra vírus Chikungunya: pontos estratégicos e desafios envolvidos nesse processo

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Abstract

Introduction: *The emergence of infections by the Chikungunya Virus (CHIKV) has been observed in different parts of the world, being a disease that can result in disabling symptoms for prolonged periods. The vaccine can be essential for controlling the disease. Vaccines must provide protection against heterologous strains, establish a correlation of protection against CHIKV infection, overcome the unpredictability of CHIKV epidemiology, and compete with other diseases for attention and limited financial resources for diseases prevalent in poor regions.* ***Objective:*** *this study identifies the profile of clinical trials related to the development of the vaccine against CHIKV and discusses strategic points and challenges involved in this process.* ***Methods:*** *the extraction of information from the selected studies was based on: authors, year of publication, study phase, type of vaccine, methodological design, number of patients, intervention protocol and outcome.* ***Results:*** *six articles representing the study of four potential vaccines were selected an attenuated virus vaccine, a nucleic acid vaccine (mRNA), a Virus Like Particle (VLP) vaccine and a viral vector vaccine. All vaccines showed good results, but unpredictable epidemiology, incorrect diagnoses and difficulty in identifying attractive markets for a CHIKV vaccine make research difficult.* ***Conclusion:*** *In recent years considerable progress has been made in research and development of vaccines against CHIKV. A variety of approaches have produced many possibilities, some of which have entered Phase I and Phase II clinical trials. However, relevant challenges such as lower economic interest in this vaccine remain.*

Keywords: *Chikungunya virus, CHIKV, Vaccines, Clinical trial*

Resumo

Introdução: *tem se observado o aparecimento de infecções pelo Virus Chikungunya (CHIKV) em diferentes partes do mundo, sendo uma doença que inclusive pode resultar em sintomas incapacitantes por períodos prolongados. A vacina pode ser essencial para o controle da doença. As vacinas devem fornecer proteção contra linhagens heterólogas, estabelecer uma correlação de proteção em relação a infecção por CHIKV, superar a imprevisibilidade da epidemiologia do CHIKV e disputar com outras doenças a atenção e o recurso financeiro limitado para doenças prevalentes nas regiões pobres.* ***Objetivo:*** *este estudo buscou identificar testes clínicos relacionados ao desenvolvimento da vacina contra o CHIKV e discutir pontos estratégicos e desafios envolvidos nesse processo.* ***Método:*** *a extração de informação dos estudos selecionados se baseou em: autores, ano de publicação, fase do estudo, tipo de vacina, desenho metodológico, número de pacientes, protocolo de intervenção e desfecho.* ***Resultados:*** *foram selecionados seis artigos que representavam o estudo de quatro vacinas em potencial, uma vacina de vírus atenuado, uma vacina de ácido nucleico (mRNA), uma vacina Virus Like Particle (VLP) e uma vacina com vetor viral. Todas as vacinas apresentaram boas respostas, porém a epidemiologia imprevisível, diagnósticos incorretos e dificuldade de identificar mercados atrativos para uma vacina contra CHIKV dificultam o andamento das pesquisas.* ***Conclusão:*** *um progresso considerável vem sendo alcançado na pesquisa e desenvolvimento de vacinas contra CHIKV. Abordagens variadas produziram possibilidades promissoras, sendo que algumas entraram na Fase I e Fase II dos testes clínicos. No entanto, desafios relevantes como menores interesses econômicos por essa vacina ainda permanecem.*

Palavras chave: *Vírus Chikungunya, CHIKV, Vacinas, Ensaio clínico*

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Introduction

In recent years, arboviruses such as Chikungunya have appeared in different parts of the world, mainly in the tropics of the African, Asian, and American continents[1-2]. The intense dispersal of these pathogens has intensified due to the expansion of global transport systems and ecological changes promoted by humans[3-4].

Chikungunya virus (CHIKV), first isolated in Tanzania in 1952-1953, is an RNA virus that belongs to the Alphavirus genus of the Togaviridae family and is transmitted to humans through the bite of the Aedes ssp mosquito. (*Aedes aegypti* and *Aedes albopictus*)[5].

CHIKV causes fever, arthralgia, and mild skin rashes and, despite a case fatality rate of less than 1%[5], the rate of progression to chronic joint manifestations ranges from 25.3% to 40.2%[6]. The most common chronic symptom is inflammatory arthralgia in the same joints affected during the acute stages, with some individuals developing arthritis similar to rheumatoid arthritis or psoriatic arthritis and may persist for months or a few years[7-8]. More severe cases, classified by the Ministry of Health as cases in which there is a need for hospitalization in intensive care or risk of death, occur in newborns and patients over 65 years old[9-10]. The impact on productivity loss becomes a relevant factor for this disease, since in 2016 a loss of approximately R$ 125 million was found[11]. Another factor that demonstrates this loss is related to the burden of the disease and the disability-adjusted life year (DALY); studies indicate that this loss reaches 2 DALY per patient[12].

Although CHIKV genotypes may vary, the strains are genetically related, and it is possible that the development of a vaccine could generate antibodies against all genotypes[13-14]. The attempt to develop a vaccine against CHIKV began in the 1960s, shortly after the virus was isolated[15].

Currently, with the evolution of biochemical and molecular methods, researchers have used several strategies to develop several alternatives, which can be classified as inactivated viral vaccine, subunit vaccine, live attenuated virus, chimeric vaccine, virus-like particle vaccine (VLP) and nucleic acid vaccine[16-17]. However, despite these scientific advances, difficulties are encountered in every technique during the development of new vaccines. Attenuated virus vaccines carry the risk of reversion to virulent strains, inactivated virus vaccines carry the risk of incomplete inactivation, subunit vaccines require the use of boosters and adjuvants, DNA/RNA vaccines can generate a greater number of side effects and VLP vaccines present limitations in productivity and increase in production costs.

Considering the epidemiological scenario of CHIKV, it is essential to develop alternatives to combat this emerging virus.

Objective

This study sought to identify Chikungunya virus vaccines under development, identifying the main techniques in the development of this vaccine and the similarities between clinical trial outcomes. In addition, the strategic points and challenges involved in this process were discussed.

Method

A bibliographic survey and literature review were carried out in PubMed, Scielo, and PMC databases.

There were no language or population groups restriction, however, the publication period was established between 2010 and 2020 for the selected articles. The research was carried out in March 2021 and the search strategy used the term: ((Chikungunya AND vaccine AND trial) OR (CHIKV AND vaccine AND trial)). The articles identified by the initial search strategy were submitted to a new filtering in which only the articles that presented in the title the terms "vaccine" and "Chikungunya" were selected. The highlighted articles were submitted to the inclusion and exclusion criteria.

Clinical trials, randomized clinical trial, cohort and case-control studies were included. Literature review articles, consensus of medical societies and expert opinion that did not present a specific clinical study related to the CHIKV vaccine were excluded from the selection.

The abstracts of the articles identified by the search strategy were evaluated in order to comply with the inclusion and exclusion criteria. In the impossibility of determining eligibility from the abstracts, the texts were read in full.

The extraction of information from the selected studies was based on: authors, year of publication, study phase, type of vaccine, methodological design, number of patients, intervention protocol and outcome. Thus, the studies were analyzed descriptively and the data presented and grouped based on methodological similarities.

Results

A total of 2476 articles were found. After selecting the papers by evaluating the titles and then applying the eligibility criteria, six articles were selected, as shown in Figure 1.

Of these, four potential vaccines were found, being

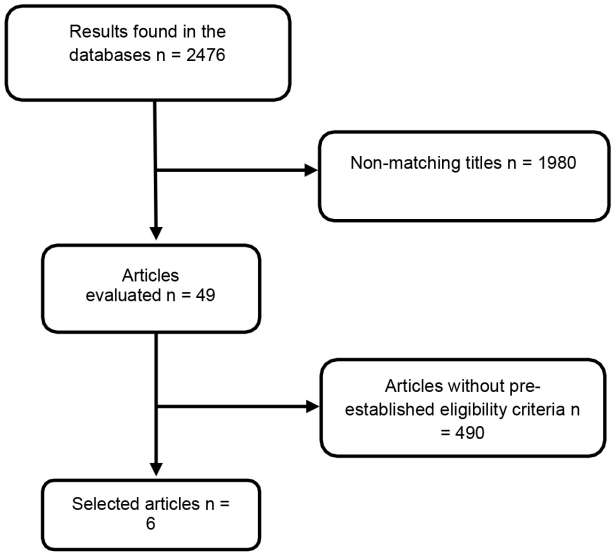

Figure 1 - Total results found and selection of articles for analysis.

an attenuated virus vaccine[18], an mRNA vaccine[19], a VLP-based vaccine[20-21] and a viral vector vaccine[22-23] with four vaccine studies in Phase I and two studies in Phase II, being five randomized and parallel studies and only one non-randomized study (Table 1).

VLA1553 Vaccine

The company Valnera (Valneva Austria GmbH, Vienna, Austria) has been developing VLA1553, an attenuated virus CHIKV vaccine candidate, with the first phase of clinical trials being carried out at two centers in the USA[24].

The study found demonstrates testing in 120 healthy volunteers aged 18 to 45 years old, who were randomly assigned to one of the escalating dose groups in a Phase I clinical trial. 31 patients received a low dose (3.2×10^3 per 0.1 mL), 30 patients received an intermediate dose (3.2×10^4 per 1 mL), and 59 patients received a high dose (3.2 × 107 [TCID50] per 1 mL). All received a single dose immunization on day 0. Subjects in all groups were revaccinated with the highest dose at month 6 or 12 and were monitored for 28 days after revaccination[18].

No vaccine-related serious adverse events were reported and data up to month 12 after a single immunization showed a good immunogenicity profile with 100% seroconversion rates. Therefore, a single dose was sufficient to induce high antibody concentrations[18].

According to ClinicalTrials.gov, in January 2022, later clinical phases studies were identified. Altogether, four Phase III clinical trials were located regarding the VLA1553 vaccine (NCT04838444, NCT04786444, NCT04546724 and NCT04650399). According to the most recent data from the website, the studies have an estimated completion date ranging between 2021 and 2025.

VAL181388 Vaccine

The development of the mRNA-based vaccine represents a partnership between Moderna (Moderna TX Inc., Massachusetts, USA) and the US government's Defense Advanced Research Projects Agency (DARPA), with clinical trials being conducted in Maryland, USA[19].

In a phase I study for mRNA-1388 (VAL181388) the candidate vaccine was tested in adults between 18 and 49 years old. A total of 60 participants were assigned to receive 25 μg (n = 15), 50 μg (n = 15) or 100 μg (n = 15) of mRNA-1388 or placebo (n = 15). Intramuscular injections were administered at weeks 0 and 4, and subjects were monitored for 1 year after the last dose. The vaccine was well tolerated at all doses studied and a dose increase in antibody titers was observed, with a substantial increase after the second vaccination [19].

VRC-CHKVLP059-00-VP Vaccine

The development of the VLP-based vaccine was carried out by the US government's National Institute of Allergy and Infectious Diseases (NIAID). Phase I was completed in the USA while phase II was carried out in the Dominican Republic, Guadeloupe, Haiti, Martinique, and Puerto Rico[25-26].

This vaccine was produced by transfecting human embryonic kidney cells (VRC293) with plasmid DNA

Table 1

CHIKV vaccine candidates in clinical development.

Strategy	*Name*	*Phase*	*Reference*
Attenuated Virus	VLA1553	I	(18)
mRNA	VAL181388	I	(19)
VLP	VRC-CHKVLP059-00-VP	I and II	(20-21)
Viral Vector	MV-CHIKV	I and II	(22-23)

expressing the structural genes of CHIKV. In this case, the VLP was evaluated in the Phase I clinical trial in 25 adults aged 18 to 50 years old. The patients were divided into 3 groups, and doses of 10 μg, 20 μg and 40 μg were administered intramuscularly at weeks 0, 4 and 20[20].

The VRC-CHKVLP059-00-VP vaccine was well tolerated and no serious adverse events were reported. 92% of participants showed induction of neutralizing antibodies after the first vaccination and all participants had neutralizing antibodies 4 weeks after the second vaccination[20].

VRC-CHKVLP059-00-VP entered a Phase II trial in 2015 in a multicenter study to evaluate safety and immunogenicity using two doses of the vaccine in 400 healthy adults between 18 and 60 years old. In this study, participants were randomized 1:1 to receive two doses of 20 μg (n = 201) or placebo (n = 199) 28 days apart and were monitored for 72 weeks. The durability of the immune response was demonstrated at the end of the study observation period[21].

MV-CHIKV Vaccine

MV-CHIKV vaccine is a viral vector vaccine using measles virus. Themis (Themis Bioscience GmbH, Wien, Austria), a subsidiary of MSD (Merck Sharp & Dohme Corp, New Jersey, USA), is responsible for the research and its clinical trials were carried out in Austria and Germany[27].

Phase I of this vaccine included 42 participants divided into four groups, being divided into low, intermediate, high, and control doses. In the control group, another viral vector vaccine that also uses the measles virus was used. The seroconversion at the first dose was 44%, 92%, and 90% for the respective groups. After a single immunization, the PRNT50 neutralizing antibody titers were 10 for the low dose, 48 for the intermediate dose and 46 for the high dose. Meanwhile, the control group revealed a titer of 7. The booster, with a 28-day interval, produced higher antibody titers in the high-dose group. To achieve 100% seroconversion, a booster dose of the vaccine was required. Overall, the vaccine showed a good safety profile, with no serious adverse events[22].

The subsequent Phase II study of the MV-CHIKV vaccine was completed in 2018. A total of 263 participants were recruited to evaluate vaccination with low or high doses at short and long intervals between first and second doses.

The results showed that the vaccine induced higher titers when given a high dose in a short interval. The results demonstrated excellent safety and tolerability[23].

Discussion

There are no specific drugs for the treatment of infections caused by CHIKV, which reveals the importance of identifying an efficient way to combat this virus. Thus, exploring the potential of different techniques for vaccine development becomes essential.

Regarding the review of vaccines for CHIKV, the attenuated virus vaccine presents the same antigens as the original pathogen, so healthy individuals develop immune responses similar to those induced by the natural infection[28]. Consequently, these vaccines induce significant responses and often confer long-term immunity after one or two doses[29].

This type of vaccine has some limitations, although to a lesser extent, clinical disease may occur after vaccination, but vaccine-induced symptoms are usually milder than after natural infection[30]. However, this type of vaccine is often contraindicated in individuals with immunodeficiency or during pregnancy. Another important point is the possibility of the attenuated virus reverting to a form capable of causing the disease[31].

When searching for Phase II and III studies with this potential vaccine, no matches were found. We believe that the appearance of SARS-CoV-2 in early 2020 caused the project to be delayed, as the company ModernaTX, Inc. possibly redirected resources towards the development of a vaccine against COVID-19. Thus, projects involving neglected diseases such as Chikungunya fever lose technical and financial resources, which promotes the postponement of research.

Unlike attenuated virus vaccines, a VLP-based vaccine requires the use of an adjuvant for sufficient long-term protection[32]. Therefore, VLP-based vaccines are safe and strongly immunogenic, however, multiple administrations with adjuvant may be required to induce complete immunity[33]. Therefore, although other studies investigating the efficacy of non-adjuvanted VLP vaccines may suggest that the single dose may be sufficient [20,34], this should be confirmed for VLPs of recombinant CHIKV in further clinical studies.

The main goals of incorporating adjuvants in vaccines are to increase immunogenicity and reduce the number of immunizations to achieve sufficient antibody titers[32].

However, depending on what is used as an adjuvant, it may increase reactogenicity and impair the vaccine's tolerability profile[35].

From the data revealed for the VLP, further studies of VRC-CHKVLP059-00-VP should identify the long-term safety of the vaccine, as the analysis during Phase II ended at week 72. Other important factors to be studied are the effectiveness and the correlation of protection of the vaccine. It is believed that the

emergence of SARS-CoV-2, in early 2020, may also have negatively interfered with the progress of new tests and Phase III, since resources were directed to the control of COVID-19.

Another promising vaccine uses the measles virus as a viral vector. Given the long-lasting response achieved by the measles vaccine, vaccines based on measles viral vectors, such as MV-CHICKV, can be considered suitable for long-term protective mass immunization[36]. Based on this, it has been hypothesized that immunity from previous measles virus infection or vaccination may interfere with protective efficacy, so this is a particular concern for this type of vaccine[37]. However, large-scale human clinical trials have demonstrated an increase in the production of measles antibodies after revaccination of previously immunized individuals[36,38].

When searching for studies on the ClinicalTrials.com platform, in December 2021, we found three more Phase II clinical studies involving this vaccine (NCT03101111, NCT03635086 and NCT03807843). This reveals that researchers are looking for more robust data to help advance clinical research to Phase III. Thus, the potential MV-CHIKV vaccine, despite showing good results in the presented Phase I and Phase II studies, needs more data to advance to Phase III of the clinical studies.

An important consideration for vaccine approaches is their ability to provide protection against heterologous strains. CHIKV has the ability to evolve into new variants in a short period of time upon entering a population, as observed in the Americas[39]. However, CHIKV still maintains a high percentage of amino acid compatibility, maintaining 95 to 99.9% in structural proteins, which implies limited diversity among CHIKV isolates[40].

Despite these gene sequence changes, studies in mice and monkeys reveal that vaccines based on a specific strain can provide long-lasting cross-protection against different strains[41]. Thus, the results indicate that a single vaccine may be able to promote protection against several strains of CHIKV.

Another important challenge for vaccine development is to establish a correlate of protection against CHIKV infection. The lack of a reliable protective ratio becomes a major obstacle to vaccine development[42], as they become more dependent on expensive and complex Phase III studies as these studies evaluate vaccine efficacy.

A factor that can collaborate with the development and licensing of vaccines for CHIKV is related to the presence of antibodies, demonstrating the importance of antibodies with neutralizing capacity for the control of infection and reinfection by CHIKV[43-44]. Another point is that the level of neutralizing antibodies showed, in animal models, a correlation with resistance to infectious challenge[33].

Obtaining reliable information about the immunological protection relationship becomes particularly important in the use of animal data, in addition to more complex human trials, which are usually requested for approval from regulatory agencies such as FDA and ANVISA (Brazilian Health Regulatory Agency).

Despite the importance of investigating vaccine safety and efficacy, the process to collect this information is not simple. The difficulty is associated with the fact that CHIKV has a highly unpredictable epidemiology, with rapid and unexpected movements that affect large populations to be followed by years of relative infectious silence[45]. In addition, many affected areas in tropical and subtropical regions of the world may not have a sophisticated system for notification, diagnosis, or surveillance of the disease. In most of these countries, other diseases such as dengue and malaria, which have similar symptoms, also circulate in the same environment, which can lead to misdiagnosis[46]. In addition, the notification of cases is often based on clinical diagnosis with the lack of serological confirmation.

The convergence of these factors leads to an inaccurate assessment of the incidence of the disease, making it difficult to study a clinical trial of efficacy, since it is based on observational data of the disease.

To allow for a statistically significant result, the clinical trial sample size is determined by the number of cases detected in the population. Therefore, planning and conducting controlled clinical trials to demonstrate the efficacy of anti-CHIKV vaccines is difficult to carry out.

Faced with the need to enable new clinical research on vaccines, an alternative will be to carry out more epidemiological studies to determine the interaction of the viruses transmitted by the Aedes mosquito. Therefore, epidemiological studies may contribute to the planning of new clinical trials, as well as vaccination strategies for different populations.

Over the years, the unpredictable epidemiology of the disease has directly influenced the development of vaccines against CHIKV, which seem to gain or lose strength depending on the emergence or disappearance of outbreaks. It is noted that with each new outbreak period, new techniques are employed, on the other hand, efforts tend to decrease due to the unpredictability of epidemiology, difficulty in demonstrating protective efficacy and limited availability of funding.

The development of a vaccine, from preclinical phase to registration, requires an increasing average investment of approximately 500 million dollars, and can exceed up to 900 million dollars[47-48], revealing that a considerable commitment from private institutions,

non-profit institutions and government institutions is required. In order to attract the resources needed to put a CHIKV vaccine into use, the commercial potential or public need must be relevant and associated with adequate risk-return criteria.

Considering that distribution and interest in the virus occur disproportionately among developed, emerging, and underdeveloped nations, industries may be unable to predict the return on investment required and thus discourage research into new vaccines. Identifying target populations for vaccination is critical to discovering the potential benefits of the vaccine in terms of return on investment. Due to its epidemic pattern and low case fatality rates, it is difficult to identify attractive markets for a CHIKV vaccine, which discourages private sector investment.

However, the unpredictability factors that drive the emergence and spread of chikungunya fever can influence the appearance of large outbreaks or that the geographic regions of the disease change, so a new market potential can be born. For better-known markets, the availability of a vaccine against Chikungunya would be a useful tool to protect local economies in endemic areas, thus preventing part of the population from being disabled for a long period.

In addition, there may also be market potential among travelers from countries that do not have recorded cases of the disease. Finally, government needs can also drive vaccine development, so in the absence of demand for the private sector, governments can fund clinical advancement.

According to data found in the literature, research on vaccines against CHIKV has progressed slowly and several possible vaccines are being available to be tested in humans. However, technical problems and financial constraints may pose potential obstacles to the development and licensing of safe and effective vaccines.

Conclusion

In recent years, considerable progress has been made in the research and development of vaccines against CHIKV. A variety of approaches have produced many possibilities, some of which have entered Phase I and Phase II clinical trials. However, relevant challenges such as lower economic interest in this vaccine remain.

Authors' contribution: The authors declare that they had equal participation in the work.

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References

1. Halstead SB. Travelling arboviruses: a historical perspective. Travel Med Infect Dis. 2019; 1:101471.
2. Ferreira AG, Fairlie S, Moreira LA. Insect vectors endosymbionts as solutions against diseases. Curr Opin Insect Sci. 2020; 40:56-61.
3. Martens P. How will climate change affect human health? the question poses a huge challenge to scientists. yet the consequences of global warming of public health remain largely unexplored. Am Sci.1999; 87(6):534-41.
4. Tatem AJ, Rogers DJ, Hay SI. Global transport networks and infectious disease spread. Adv Parasitol. 2006; 62:293-343.
5. Cunha MS, Costa PAG, Correa IA, de Souza MRM, Calil PT, da Silva GPD, et al. Chikungunya virus: an emergent arbovirus to the south American continent and a continuous threat to the world. Front Microbiol. 2020; 11:1297.
6. Rodriguez-Morales AJ, Cardona-Ospina JA, Urbano-Garzon SF, Hurtado-Zapata JS. Prevalence of post-chikungunya chronic inflammatory rheumatism: a systematic review and meta-analysis. Arthritis Care Res (Hoboken). 2016; 68:1849-58.
7. Thiberville SD, Boisson V, Gaudart J, Simon F, Flahault A, de Lamballerie X. Chikungunya fever: a clinical and virological investigation of outpatients on Reunion Island, South-West Indian Ocean. PLoS Negl Trop Dis. 2013;7(1):e2004.
8. Roosenhoff R, Anfasa F, Martina B. The pathogenesis of chronic chikungunya: evolving concepts. Fut Virol. 2015; 11(1):61-77.
9. Brasil. Ministério da Saúde. Secretaria de Vigilância em Saúde. Secretaria de Atenção Básica. Febre de chikungunya: manejo clínico. Brasília(DF): Ministério da Saúde; 2015. 28p.
10. Morrison TE. Reemergence of chikungunya virus. J Virol. 2014; 88(20):11644-7.
11. Teich V, Arinelli R, Fahham L. Aedes aegypti and society: the economic burden of arboviruses in Brazil. J Bras Econ Saúde 2017; 9(3):267-76.
12. Puntasecca CJ, King CH, LaBeaud AD. Measuring the global burden of chikungunya and Zika viruses: A systematic review. PLoS Negl Trop Dis. 2021; 15(3):e0009055.
13. Harrison VR, Binn LN, Randall R. Comparative immunogenicities of chikungunya vaccines prepared in avian and mammalian tissues. Am J Trop Med Hyg. 1967; 16(6):786-91.
14. Goo L, Dowd KA, Lin TY, Mascola JR, Graham BS, Ledgerwood JE, et al. A virus-like particle vaccine elicits broad neutralizing antibody responses in humans to all chikungunya virus genotypes. J Infect Dis. 2016; 214(10):1487-91.
15. Powers AM. Vaccine and therapeutic options to control chikungunya virus. Clin Microbiol Rev. 2017; 31(1):e00104-16.
16. Weaver SC, Osorio JE, Livengood JA, Chen R, Stinchcomb DT. Chikungunya virus and prospects for a vaccine. Expert Rev Vaccines. 2012; 11(9):1087-101.
17. Mao HH, Chao S. Advances in vaccines. Adv Biochem Eng Biotechnol. 2020; 171:155-88.
18. Wressnigg N, Hochreiter R, Zoihsl O, Fritzer A, Bézay N, Klingler A, et al. Single-shot live-attenuated chikungunya vaccine in healthy adults: a phase 1, randomised controlled trial. Lancet Infect Dis. 2020; 20(10):1193-203.
19. Shaw C, Panther L, August A, Zaks T, Smolenov I, Bart S, Watson M. Safety and immunogenicity of a mRNA-based chikungunya vaccine in a phase 1 dose-ranging trial. Int J Infect Dis. 2019; 79(S1):17.
20. Chang LJ, Dowd KA, Mendoza FH, Saunders JG, Sitar S, Plummer SH, et al. Safety and tolerability of chikungunya virus-like particle vaccine in healthy adults: a phase 1 dose-escalation trial. Lancet. 2014; 384(9959):2046-52.
21. Chen GL, Coates EE, Plummer SH, Carter CA, Berkowitz N, Conan-Cibotti M, et al. Effect of a chikungunya virus-

like particle vaccine on safety and tolerability outcomes: a randomized clinical trial. JAMA. 2020; 323(14):1369-77.
22. Ramsauer K, Schwameis M, Firbas C, Müllner M, Putnak RJ, Thomas SJ, et al. Immunogenicity, safety, and tolerability of a recombinant measles-virus-based chikungunya vaccine: a randomised, double-blind, placebocontrolled, active-comparator, first-in-man trial. Lancet Infect Dis. 2015;15(5):519-27.
23. Reisinger EC, Tschismarov R, Beubler E, Wiedermann U, Firbas C, Loebermann M, et al. Immunogenicity, safety, and tolerability of the measles-vectored chikungunya virus vaccine MV-CHIK: a double-blind, randomised, placebo-controlled and active-controlled phase 2 trial. Lancet. 2019; 392(10165):2718-27.
24. U.S. National Library of Medicine. ClinicalTrials.gov. Study to assess the safety and immunogenicity of a chikungunya virus vaccine candidate (VLA1553) in healthy volunteers. ClinicalTrials.gov Identifier: NCT03382964. August 29, 2019 [Internet]. Bethesta (MD): National Library of Medicine; 2019. [citado 2021 Dec 20]. Disponível em: https://clinicaltrials.gov/ct2/show/NCT03382964
25. U.S. National Library of Medicine. ClinicalTrials.gov. Chikungunya virus vaccine trial in healthy adults. ClinicalTrials.gov Identifier: NCT01489358. July 25, 2016 [Internet]. Bethesda (MD): National Library of Medicine, 2016. [citado 2021 Dec 20]. Disponível em: https://clinicaltrials.gov/ct2/show/NCT01489358
26. U.S. National Library of Medicine. ClinicalTrials.gov. Trial for safety and immunogenicity of a chikungunya vaccine, VRC-CHKVLP059-00-VP, in healthy adults. ClinicalTrials.gov Identifier: NCT02562482. October 22, 2020. [Internet]. Bethesda (MD): National Library of Medicine; 2020. [citado 2021 Dec 20]. Disponível em: https://clinicaltrials.gov/ct2/show/NCT02562482
27. U.S. National Library of Medicine. ClinicalTrials.gov. Phase II study to evaluate safety and immunogenicity of a chikungunya vaccine (MV-CHIK-202). ClinicalTrials.gov Identifier: NCT02861586. October 22, 2021. [Internet]. Bethesda (MD): National Library of Medicine; 2021. [citado 2021 Dec 20]. Disponível em: https://clinicaltrials.gov/ct2/show/NCT02861586
28. Plotkin SA, Orenstein WA, Offit PA. Vaccines. 5th ed. Philadelphia: Saunders; 2008. 1748p.
29. Pulendran B, Ahmed R. Immunological mechanisms of vaccination. Nat Immunol. 2011;12(6):509-17.
30. Fuenmayor J, Gòdia F, Cervera L. Production of virus-like particles for vaccines. N Biotechnol. 2017; 39(Pt B):174-80.
31. Minor PD. Live attenuated vaccines: historical successes and current challenges. Virology. 2015; 479-480:379-92.
32. Cimica V, Galarza JM. Adjuvant formulations for virus-like particle (VLP) based vaccines. Clin Immunol. 2017; 183:99-108.
33. Akahata W, Yang ZY, Andersen H, Sun S, Holdaway HA, Kong WP, et al. A virus-like particle vaccine for epidemic chikungunya virus protects nonhuman primates against infection. Nature Med. 2010; 16(3):334–9.
34. Metz SW, Gardner J, Geertsema C, Le TT, Goh L, Vlak JM, et al. Effective chikungunya virus-like particle vaccine produced in insect cells. PLoS Negl Trop Dis. 2013;7:e2124.
35. Petrovsky N, Aguilar JC. Vaccine adjuvants: current state and future trends. Immunol Cell Biol. 2004; 82(5):488-96.
36. Wong-Chew RM, Beeler JA, Audet S, Santos JI. Cellular and humoral immune responses to measles in immune adults re-immunized with measles vaccine. J Med Virol. 2003; 70(2):276-80.
37. Mühlebach MD. Vaccine platform recombinant measles virus. Virus Genes. 2017; 53(5):733-40.
38. Rager-Zisman B, Bazarsky E, Skibin A, Chamney S, Belmaker I, Shai I, et al. The effect of measles-mumpsrubella (MMR) immunization on the immune responses of previously immunized primary school children. Vaccine. 2003; 21(19-20):2580-8.
39. Sahadeo NSD, Allicock OM, De Salazar PM, Auguste AJ, Widen S, Olowokure B, et al. Understanding the evolution and spread of chikungunya virus in the Americas using complete genome sequences. Virus Evol. 2017; 3(1):vex010.
40. Stapleford KA, Moratorio G, Henningsson R, Chen R, Matheus S, Enfissi A, et al. Whole-genome sequencing analysis from the chikungunya virus caribbean outbreak reveals novel evolutionary genomic elements. PLoS Negl Trop Dis. 2016;10:e0004402.
41. Langsjoen RM, Haller SL, Roy CJ, Vinet-Oliphant H, Bergren NA, Erasmus JH, et al. Chikungunya virus strains show lineage-specific variations in virulence and cross-protective ability in murine and nonhuman primate models. mBio. 2018; 9(2):e02449-17.
42. Bhatt K, Verma S, Ellner JJ, Salgame P. Quest for correlates of protection against tuberculosis. Clin Vaccine Immunol. 2015; 22(3):258-66.
43. Kam YW, Lum FM, Teo TH, Lee WW, Simarmata D, Harjanto S, et al. Early neutralizing IgG response to Chikungunya virus in infected patients targets a dominant linear epitope on the E2 glycoprotein. EMBO Mol Med. 2012; 4(4):330-43.
44. Couderc T, Khandoudi N, Grandadam M, Visse C, Gangneux N, Bagot S, et al. Prophylaxis and therapy for Chikungunya virus infection. J Infect Dis. 2009; 200(4):516–23.
45. Wahid B, Ali A, Rafique S, Idrees M. Global expansion of chikungunya virus: mapping the 64-year history. Int J Infect Dis. 2017; 58:69-76. Epub 2017 Mar 10.
46. Alvarado LI, Lorenzi OD, Torres-Velásquez BC, Sharp TM, Vargas L, Muñoz-Jordán JL, et al. Distinguishing patients with laboratory-confirmed chikungunya from dengue and other acute febrile illnesses, Puerto Rico, 2012-2015. PLoS Negl Trop Dis. 2019; 13(7):e0007562.
47. Andre FE. How the research-based industry approaches vaccine development and establishes priorities. Dev Biol (Basel). 2002; 110:25–9.
48. Pronker ES, Weenen TC, Commandeur H, Claassen EH, Osterhaus AD. Risk in vaccine research and development quantified. PLoS One. 2013; 8(3):e57755.

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