VACCINATION RESEARCH

Openventio PUBLISHERS

Open Journal

http://dx.doi.org/10.17140/VROJ-1-106

Review

*Corresponding author
Sajani Dias, PhD
School of Science, BMS
No. 591 Galle Rd
Colombo 6, Sri Lanka
Tel. +94 72 7001087
E-mail: biomedical@bms.lk

Volume 1 : Issue 1
Article Ref. #: 1000VROJ1106

Article History
Received: May 25th, 2017
Accepted: May 31st, 2017
Published: May 31st, 2017

Citation
Sameem R, Dias S. Ebola virus: Promising vaccine candidates. *Vaccin Res Open J*. 2017; 1(1): 33-38. doi: 10.17140/VROJ-1-106

Ebola Virus: Promising Vaccine Candidates

Reema Sameem, Bsc (Hons); Sajani Dias, PhD*

School of Science, BMS, No. 591 Galle Rd, Colombo 6, Sri Lanka

ABSTRACT

Ebola Virus Disease is among the deadliest viral diseases. The field of Ebola vaccine development has progressed over the years with numerous candidates in advanced stages of clinical development. Currently, there is no licensed vaccine against Ebola virus. This review aimed to discuss the promising Ebola vaccine candidates focusing on vaccines against ebola viruses in particular against Zaire ebola virus (ZEBOV). Although DNA vaccines have been evaluated in Phase I clinical studies, it demonstrates low immunogenicity. Thus far, the most successful vaccine platforms are the recombinant viruses including, replicating vesicular stomatitis virus (rVSV) and chimpanzee adenovirus 3 (ChAd3). The successful trials attests that rVSV vaccines will be submitted for licensing in the near future.

KEY WORDS: Ebola virus; ZEBOV; rVSV; ChAd3; Vaccine; rAd5; Immunization; Clinical trials.

ABBREVIATIONS: ZEBOV: Zaire ebola virus; rVSV: replicating vesicular stomatitis viru; ChAd3: Chimpanzee adenovirus 3; EVD: Ebola Virus Disease; CIEBOV: *Cote d'Ivoire Ebolavirus*; REBOV: *Reston Ebolavirus.*

INTRODUCTION

Ebola virus disease (EVD) is among the most virulent viral infections caused by the Ebola virus, with mortality rates nearing 90%.[1,2] Ebola virus is a member of the *Filoviridae* family, and is classified into five species, *Zaire Ebolavirus* (ZEBOV), *Sudan Ebolavirus* (SEBOV), *Bundibugyo Ebolavirus* (BEBOV), *Cote d'Ivoire Ebolavirus* (CIEBOV) and *Reston Ebolavirus* (REBOV) in decreasing order of virulence.[3,4]

Ebola virus particularly, SEBOV and ZEBOV were discovered in 1976 in the Democratic Republic of Congo. Since then the virus has initiated over 20 sporadic outbreaks restricted to regions of Africa.[5,6] Consequently, in 2014, the World Health Organization (WHO) declared an urgent need for efficacy and safety testing of EVD vaccine candidates.[7]

The largest EVD epidemic to date, was reported between December 2013 and April 2016, which generated over 28,000 cases and 11,000 deaths in the African populations of Guinea, Liberia and Sierra Leone.[1,8] Up-to-date, there are no reported cases of EVD in Sri Lanka. However, due to the inevitable re-emergence of the disease, WHO has warned all countries to intensify surveillance activities to combat any possible threat of EVD. Therefore, Sri Lanka has strengthened its scrutiny activities in order to recognize, notify and manage suspected EVD patients.

The devastating effects of the 2014 EVD outbreak spurred international research into vaccine development. Although, it has been 40 years since the discovery of Ebola virus, there are no approved vaccines for the disease. Therefore, there is an urgent need for an Ebola vaccine which is cheap, effective and safe, which can be administered in a single dose.[9,10] The aim of this review is to discuss the most promising candidate vaccines which could potentially protect against the Ebola virus.

Openventio PUBLISHERS
http://dx.doi.org/10.17140/VROJ-1-106

Ebola Virus Genome

Ebola virus genome is the prime target for potential candidate vaccines. Ebola virus is a single-stranded, lipid enveloped, negative sense RNA virus of approximately 19 kb, which comprises of genes that code for seven proteins.[11] Four of the genes encode structural proteins including nucleoprotein (NP), glycoprotein (GP) and two viral matrix proteins VP24 and VP40. Whereas, VP30, VP35 and the RNA-dependent viral polymerase (L) are the non-structural proteins.[12,13] Each gene encodes functional proteins for viral replication.

Moreover, it has been recorded that vaccines for the protection against EVD predominantly target glycoprotein, with an exemption of a few directed at VP40 and nucleoprotein.[14-16]

STRATEGIES FOR EBOLA VACCINE DEVELOPMENT

The recent Ebola outbreak brought global attention to the inadequate preventive and therapeutic measures against EVD. This intern spurred intense action towards initiation and development of an Ebola vaccine. The first successful Ebola virus vaccine was a conventional inactivated vaccine developed by Lupton and colleagues in 1980.[17,18] However, due to the possible threat of incomplete inactivation *versus* using the term reversion to virulence, inactivated immunizations were not encouraged against EVD.

Over the past two decades, researchers have explored various strategies for Ebola vaccine development. Broadly, Ebola vaccine candidates can be categorised into replication incompetent and replication competent vaccines.[19,20] In the following subsections, the three most promising vaccine candidates currently undergoing clinical trials are discussed.

Replication Incompetent (Non-Replicative) Vaccines

Recombinant adenovirus based vaccines: Adenoviruses are double-stranded, non-enveloped DNA viruses, which have been isolated from mammalian species. Adenoviruses are used as recombinant vectors by deletion of the E1 region, thus making the virus replicative deficient.[21,22] Various clinical trials have demonstrated the safety of adenovirus vectors for use as a benign carrier system. In 2000, Sullivan and fellow scientists, first documented the use of recombinant Adenovirus 5 (rAd5) – based vectors which expressed EBOV antigens as a vaccine.[23]

Majority of the first generation adenovirus vectors focused on the innovation of a vaccine based on human serotype (AdHu5). However, pre-existing immunity greatly decline the efficacy of AdHu5 immunizations.[24] Several strategies have been derived to overcome pre-existing immunity, including the use of a variety of serotypes of recombinant Adenovirus such as the rare human serotypes Ad26 and Ad35. Subsequently, the second generation of adenovirus was developed from Chimpanzee Adenovirus, Ad3, Ad7 and Ad62.[25]

Initially, rAd5 vaccines were used to boost immunizations upon priming them with DNA vaccines. Although, the vaccination strategy provided 100% protection to Non-human primates (NHP) against the ZEBOV challenge, a period of over 6 months was required to complete immunization.[26,27] This study was found to be highly effective and dependent of Ebola-specific CD8+ T-cells and antibody responses. Moreover, CD8+ cells played a significant role in rAd5-GP-induced immunity against EBOV infection in NHPs.[28,29] Additionally, vaccination dose is a crucial factor in determining vaccine efficacy. Studies demonstrated that at least 1×10^{10} virus particles are necessary to attain 100% protection in NHPs against Ebola virus infections.[30,31]

In 2010, a double-blinded, placebo-controlled, dose-escalation, Phase I human study confirmed the safety and immunogenicity of rAd5 immunization encoding the envelope GP from ZEBOV and SEBOV 1976 strain. Thirty-one healthy adults were vaccinated at 2×10^{9} (n=12), or 2×10^{10} (n=11) viral particles or placebo (n=8) as an intramuscular injection. All participants demonstrated antigen specific humoral and cellular immune responses.[32]

In 2015, a placebo-controlled, double-blind, phase 1 clinical trial was performed in China, to evaluate the safety and immunogenicity of rAd5 vaccine encoding the envelope GP-ZEBOV 2014 strain. In this study, 120 healthy adults were randomly allocated to receive placebo (n=40), low-dose (n=40) or high-dose (n=40) vaccine. The results of the study demonstrated that the high-dose immunization is safe and robustly immunogenic. Single inoculation of the high-dose vaccine could mount glycoprotein-specific humoral and T-cell response against Ebola virus in fourteen days.[33]

Subsequent studies were carried out to test the rare serotype rAd vectors to circumvent Ad5 immunity. The results demonstrated that the great extent of CD8+ T-cell responses were not constantly prognostic of vaccine efficacy. In 2011, Geisbert and team identified that Macaques immunized with a single dose of rAd26 and rAd35 EBOV vaccines at the uniformly protective rAd5 vaccine dose, 10^{10} particles, produced serum anti-glycoprotein Abs along with T-cell responses of CD4+ and CD8+ comparable to rAd5. Nonetheless, this was unsuccessful in attempts to protect animals against EBOV infection.[34]

Evaluation of the cytokine secretion patterns within antigen-specific T-cells following Ebola immunization exhibited a prominent difference between rAd5 and the rare serotype rAd26 or rAd35 inoculations. It was found that the CD8+ T-cell response initiated by the protective rAd5 vaccine was significantly greater compared to the rarer serotypes, and consisted predominantly of TNF/IFN-γ double positive, cytolytic effector cells.[35]

Recently, Ad26 vaccine expressing the full-length GP of ZEBOV was administered in a prime-boost regimen with multivalent Modified Vaccinia Ankara (MVA)-Bavarian Nordic (BN) vaccine, which is currently undergoing Phase III trials.[36,37]

Openventio PUBLISHERS
http://dx.doi.org/10.17140/VROJ-1-106

Candidate vaccines based on the chimpanzee adenovirus 3 (ChAd3) platform are currently being developed by deletion and replacement of the E1 gene in the cAd3 genome with EBOV GP. Between September and December 2014, five phase 1 trials of ChAd3 commenced in North America, Europe and Africa.[38,39]

The preliminary report of the first ChAd3 trial, confirms the safety and immunogenicity of the vaccine. In this study, twenty healthy adults were enrolled in groups of 10 each, and were subjected to intramuscular immunization in doses of 2×10^{10} or 2×10^{11} particle units. All 20 participants produced Glycoprotein-specific antibodies and the 2×10^{11} particle unit dose group had a greater magnitude titer than 2×10^{10} particle unit dose group. Therefore, the immune responses and reactogenicity to cAd3-EBO vaccine were dose-dependent.[40,41]

Phase II and III trials were initiated in Liberia, Sierra Leone and Guinea. In February 2015, the trial known as Partnership for research on Ebola vaccines in Liberia (PREVAIL) was initiated.[41,42]

Furthermore, analysis of Ebola virus vaccine vectors demonstrated that robust antibody titers and high magnitude of TNF/IFN-γ double positive CD8+ T-cell quality in ChAd3 are remarkably similar to rAd5 in inoculated macaques.[43]

In 2014, researchers developed a trivalent engineered Ebola-Marburg vaccine (GreEMTri), third generational Adenovirus based vaccines, which expresses glycoprotein genes of both ZEBOV and SEBOV along with deletion of all Ad genes. Advantages of Adenovirus based vector vaccinations include, high immunogenicity, targeted immune response, multi-delivery routes and effectiveness at low doses.[44] Altogether, the available data have shown great potential for adenovirus-based vaccines to be licensed in the future.

REPLICATION COMPETENT VACCINES

Recombinant Vesicular Stomatitis Virus (rVSV)

rVSV is a member of the family *Rhabdoviridae*, and a promising vaccine platform for EBOV. In 1987, Rose et al, pioneered the use of rVSV as a vaccine vector.[45] The rVSV vector vaccine is designed to function by replacing the rVSV-G with a G from an EBOV strain by using reverse genetics.[46] This chimeric alteration attenuates the pathogenicity and the neurotropism of the rVSV delta G filovirus GP vectors for ZEBOV and MARV, while allowing the vaccine virus to replicate using the Ebola GP to attach and enter cells.[47]

In 2005, a vaccine based on rVSV was the first replicating Ebola virus vaccine shown to be protective in NHPs. In this study, a single intramuscular vaccination elicited protective immune responses in NHPs against lethal EBOV challenge. The EBOV immunization triggered humoral and cellular immune reactions in all inoculated NHPs and provided 100% protection.[48]

Furthermore, the rVSV vaccines have shown remarkable post-exposure success. In a study conducted in Canada, guinea pigs and mice were first inoculated with $2x10^4$ or 2×10^5 plaque-forming unit (PFU) of rVSV-EBOV vaccine respectively. Challenge of mice with a lethal dose of Mouse Adapted EBOV at 6.5 and 9 months after immunization proved complete protection, and at 12 months post-vaccination, 80% (12 of 15 survivors) protection. Similarly, encounter of guinea pigs with a lethal dose of guinea pig-adapted EBOV at 7, 12 and 18 months after vaccination resulted in 83% (5 of 6 survivors) at 7 months after vaccination, and 100% survival at 12 and 18 months after vaccination. The AB responses were examined using sera from each rodent. Additionally, there was a correlation between the quantity of EBOV GP-specific IgG Ab and protection. This study concluded that inoculation with rVSV-EBOV is able to confer long-term protection against EVD in guinea pigs and mice.[49,50]

Up to date, the rVSV-ZEBOV immunization has been investigated in eight human phase I trials across Europe, Africa and North America. Moreover, the Sierra Leone trial to introduce a vaccine against Ebola (STRIVE) phase III study in Sierra Leone is ongoing.[51,52]

Phase I rVSV-ZEBOV double-blind, placebo-controlled, dose-escalation trial was conducted on 52 volunteers. In this study, 12 volunteers received placebo injection and 40 participants received rVSV-ZEBOV immunization at either an intramuscular dose of 3 million PFU or 20 million PFU. The safety and immunogenicity were evaluated for 28 days post-immunization. The results demonstrated that, all participants had seroconversions by day 28, as assessed by ELISA against the GP of ZEBOV-Kikwit strain. On day 28, the group receiving 20 million PFU had a higher geometric mean titer of Ab against ZEBOV GP than the group receiving 3 million PFU. These preliminary outcomes promote the further development of the vaccine dose of 20 million PFU.[53]

A similar Phase 1/2 clinical trial in Geneva was carried out to evaluate the safety and immunogenicity of rVSV vaccine administered at different doses. In this study, 59 healthy volunteers receiving $3x10^5$ PFU low dose of rVSV-ZEBOV vaccine were compared with rVSV-ZEBOV high dose vaccines $1x10^7$ PFU (n=35) or $5x10^7$ PFU (n=16) or a placebo (n=8). Initially, viral oligoarthritis was detected in eleven of the first 51 participants (22%) subjected to receive 10^7 or 5×10^7 PFU. Thereafter, 56 volunteers received a lower dose 3×10^5 PFU (n=51) or placebo (n=5) to examine the influence of dose reduction on safety and immunogenicity. The study concluded that reduction in the dose of rVSV-ZEBOV enhanced early tolerability, nevertheless suppressed antibody responses.[54]

A Phase III efficacy trial is currently underway in Guinea to evaluate the efficacy of rVSV-ZEBOV as a preventative strategy for EVD. The trial used a ring vaccination strategy and the preliminary reports demonstrated encouraging results. In this ongoing study, 90 groups (7651 individuals) comprising

of contacts of an index case received 20 million PFU of rVSV either immediately or 21 days after documentation of the index case. In the immediate vaccination group, no cases of EVD were reported whereas 16 cases of EVD were diagnosed in the delayed vaccination group, 10 days post-vaccination. The results show a vaccine efficacy of 100%. From six days following vaccination, there were no new cases of EVD identified in vaccine recipients from the immediate or delayed groups. The data depicts that rVSV confers protection between 6-21 days after vaccination, nonetheless the longevity of the vaccine-induced protection is unknown.[55]

Advantages of using rVSV vaccines include, very limited pre-existing immunity, easily propagation in mammalian cells and capacity to initiate a strong humoral and cellular immune responses. Additionally, rVSV based vaccines have the ability to confer both systemic and mucosal immunity.[56] These clinical studies demonstrated the safety and efficacy of rVSV vaccine. Researchers predict this vaccine will be submitted for full licensing by the end of 2017.

SUMMARY

Ebola vaccine development has progressed at an exponential rate with numerous candidates in advanced stages of clinical development. Although, DNA vaccines have been evaluated in Phase I clinical studies, it demonstrates low immunogenicity and necessitates frequent vaccinations over a sustained period of time. Thereby challenging the implementation of DNA vaccine platforms as vaccination of the entire population over a long time period is logistically and financially not feasible.

Thus far, the most promising Ebola vaccine candidates are the live-replicating rVSV and the replication-defective ChAd3 based on GP of ZEBOV strain of Ebola virus. Multiple trials currently underway in Europe and Africa have demonstrated that these vaccines are safe and immunogenic. ChAd3 and Ad26 successfully passed through clinical trials and entered into a Phase III trials, whereas rVSV has been validated to be effective in a Phase III clinical studies. While the efficacy of rVSV vaccine is encouraging, challenges exist in improving vaccines to provide durable efficacy and recognizing optimal pathways for vaccine delivery. At this point, the crucial impediment for EBOV vaccines to move forward is the limited funding for vaccine development.

In summary, extensive research carried out on rVSV vaccine depicts that this persists to be the only immunization platform for prospective clinical use. Its efficacy following a single dose is a substantial benefit for providing immunizations to target populations. Moreover, this vaccine is cost-effective and is efficacious both pre- and post-exposure with a moderately short time to immunity. The next steps would be to develop a pathway to licence rVSV vaccines and potentially stockpile vaccines for future Ebola outbreaks. An effective Ebola vaccine strategy can be envisioned for use, in order to limit the spread of an outbreak and to protect individuals who are at the highest risk of infection.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest.

REFERENCES

1. Leligdowicz A, Fischer WA 2nd, Uyeki TM, et al. Ebola virus disease and critical illness. *Crit Care*. 2016; 20(1): 217. doi: 10.1186/s13054-016-1325-2

2. Wambani RJ, Ogola PE, Arika WM, Rachuonyo HO, Burugu MW. Ebola virus disease: A biological and epidemiological perspective of a virulent virus. *J Infect Dis Diagn*. 2016; 1: 103. Web site. http://ir-library.ku.ac.ke/handle/123456789/14250. Accessed May 24, 2017.

3. Nakayama E, Saijo M. Animal models for Ebola and Marburg virus infections. *Front Microbiol*. 2013; 4: 267. doi: 10.3389/fmicb.2013.00267

4. Laupland KB, Valiquette L. Ebola virus disease. *Can J Infect Dis Med Microbiol*. 2014; 25(3): 128-129.

5. Chan M. Ebola virus disease in West Africa — No early end to the outbreak. *N Engl J Med*. 2014; 371(13): 1183-1185. Web site. http://www.nejm.org/doi/pdf/10.1056/nejmp1409859. Accessed May 24, 2017.

6. Urbanowicz RA, McClure CP, Sakuntabhai A, et al. Human adaptation of Ebola virus during the West African outbreak. *Cell*. 2016; 167(4): 1079-1087e5.

7. Uyeki TM, Mehta AK, Davey RT, Jr., et al. Clinical management of Ebola virus disease in the United States and Europe. *N Engl J Med*. 2016; 374(7): 636-646. doi: 10.1016/j.cell.2016.10.013

8. Team WER. After Ebola in West Africa — Unpredictable risks, preventable epidemics. *N Engl J Med*. 2016; 375(6): 587-596. doi: 10.1056/NEJMsr1513109

9. Burghardt K, Verzijl C, Huang J, Ingram M, Song B, Hasne M-P. Testing modeling assumptions in the West Africa Ebola outbreak. *Sci Rep*. 2016; 6: 34598. doi: 10.1038/srep34598

10. Sullivan N, Yang ZY, Nabel GJ. Ebola virus pathogenesis: Implications for vaccines and therapies. *J Virol*. 2003; 77(18): 9733-9737. doi: 10.1128/JVI.77.18.9733-9737.2003

11. Lai KY, Ng WYG, Cheng FF. Human Ebola virus infection in West Africa: A review of available therapeutic agents that target different steps of the life cycle of Ebola virus. *Infect Dis Poverty*. 2014; 3(1): 43. doi: 10.1186/2049-9957-3-43

12. Trunschke M, Conrad D, Enterlein S, Olejnik J, Brauburger

K, Muhlberger E. The L-VP35 and L-L interaction domains reside in the amino terminus of the Ebola virus L protein and are potential targets for antivirals. *Virology*. 2013; 441(2): 135-145. doi: 10.1016/j.virol.2013.03.013

13. Diehl WE, Lin AE, Grubaugh ND, et al. Ebola virus glycoprotein with increased infectivity dominated the 2013-2016 Epidemic. *Cell*. 2016; 167(4): 1088-1098.e6. doi: 10.1016/j.cell.2016.10.014

14. Wilson JA, Bray M, Bakken R, Hart MK. Vaccine potential of Ebola virus VP24, VP30, VP35, and VP40 proteins. *Virology*. 2001; 286(2): 384-390. doi: 10.1006/viro.2001.1012

15. Kondratowicz AS, Maury WJ. Ebola virus: A brief review of novel therapeutic targets. *Future Microbiol*. 2012; 7(1): 1-4. doi: 10.2217/fmb.11.110

16. Mehedi M, Falzarano D, Seebach J, et al. A new Ebola virus nonstructural glycoprotein expressed through RNA editing. *J Virol*. 2011; 85(11): 5406-5414. doi: 10.1128/JVI.02190-10

17. Blaney JE, Wirblich C, Papaneri AB, et al. Inactivated or live-attenuated bivalent vaccines that confer protection against rabies and Ebola viruses. *J Virol*. 2011; 85(20): 10605-10616. doi: 10.1128/JVI.00558-11

18. Geisbert TW, Pushko P, Anderson K, Smith J, Davis KJ, Jahrling PB. Evaluation in nonhuman primates of vaccines against Ebola virus. *Emerg Infect Dis*. 2002; 8(5): 503-507. doi: 10.3201/eid0805.010284

19. Halfmann P, Ebihara H, Marzi A, et al. Replication-deficient ebolavirus as a vaccine candidate. *J Virol*. 2009; 83(8): 3810-3815. doi: 10.1128/JVI.00074-09

20. Robert-Guroff M. Replicating and non-replicating viral vectors for vaccine development. *Curr Opin Biotechnol*. 2007; 18(6): 546-556. doi: 10.1016/j.copbio.2007.10.010

21. Ura T, Okuda K, Shimada M. Developments in viral vector-based vaccines. *Vaccines*. 2014; 2(3): 624-641. doi: 10.3390/vaccines2030624

22. Dudek T, Knipe DM. Replication-defective viruses as vaccines and vaccine vectors. *Virology*. 2006; 344(1): 230-239. doi: 10.1016/j.virol.2005.09.020

23. Sullivan NJ, Sanchez A, Rollin PE, Yang ZY, Nabel GJ. Development of a preventive vaccine for Ebola virus infection in primates. *Nature*. 2000; 408(6812): 605-609. doi: 10.1038/35046108

24. Mast TC, Kierstead L, Gupta SB, et al. International epidemiology of human pre-existing adenovirus (Ad) type-5, type-6, type-26 and type-36 neutralizing antibodies: Correlates of high Ad5 titers and implications for potential HIV vaccine trials. *Vaccine*. 2010; 28(4): 950-957. doi: 10.1016/j.vaccine.2009.10.145

25. Wu S, Kroeker A, Wong G, et al. An adenovirus vaccine expressing Ebola virus variant makona glycoprotein is efficacious in guinea pigs and nonhuman primates. *J Infect Dis*. 2016; 214(suppl 3): S326-S332. doi: 10.1093/infdis/jiw250

26. Stanley DA, Honko AN, Asiedu C, et al. Chimpanzee adenovirus vaccine generates acute and durable protective immunity against ebolavirus challenge. *Nat Med*. 2014; 20(10): 1126-1129. doi: 10.1038/nm.3702

27. Quinn KM, Zak DE, Costa A, et al. Antigen expression determines adenoviral vaccine potency independent of IFN and STING signaling. *J Clin Invest*. 2015; 125(3): 1129-1146. doi: 10.1172/JCI78280

28. Wong G, Richardson JS, Pillet S, et al. Immune parameters correlate with protection against ebola virus infection in rodents and nonhuman primates. *Sci Transl Med*. 2012; 4(158): 158ra146. doi: 10.1126/scitranslmed.3004582

29. O'Brien LM, Stokes MG, Lonsdale SG, et al. Vaccination with recombinant adenoviruses expressing Ebola virus glycoprotein elicits protection in the interferon alpha/beta receptor knock-out mouse. *Virology*. 2014; 452-453: 324-333. doi: 10.1016/j.virol.2013.03.028

30. Hensley LE, Mulangu S, Asiedu C, et al. Demonstration of cross-protective vaccine immunity against an emerging pathogenic Ebolavirus Species. *PLoS Pathog*. 2010; 6(5): e1000904. doi: 10.1371/journal.ppat.1000904

31. Sridhar S. Clinical development of Ebola vaccines. *Ther Adv Vaccines*. 2015; 3(5-6): 125-138. doi: 10.1177/2051013615611017

32. Ledgerwood JE, Costner P, Desai N, et al. A replication defective recombinant Ad5 vaccine expressing Ebola virus GP is safe and immunogenic in healthy adults. *Vaccine*. 2010; 29(2): 304-313. doi: 10.1016/j.vaccine.2010.10.037

33. Zhu FC, Hou LH, Li JX, et al. Safety and immunogenicity of a novel recombinant adenovirus type-5 vector-based Ebola vaccine in healthy adults in China: Preliminary report of a randomised, double-blind, placebo-controlled, phase 1 trial. *Lancet*. 2015; 385(9984): 2272-2279. doi: 10.1016/S0140-6736(15)60553-0

34. Geisbert TW, Bailey M, Hensley L, et al. Recombinant adenovirus serotype 26 (Ad26) and Ad35 vaccine vectors bypass immunity to Ad5 and protect nonhuman primates against ebolavirus challenge. *J Virol*. 2011; 85(9): 4222-4233. doi: 10.1128/JVI.02407-10

35. Colloca S, Folgori A, Ammendola V, et al. Generation and screening of a large collection of novel simian Adenovirus allows the identification of vaccine vectors inducing potent cellular immunity in humans: A range of novel simian adenoviral vectors, which are capable of priming high levels of T cell responses in man, has been defined. *Sci Transl Med*. 2012; 4(115): 115ra2-115ra2. doi: 10.1126/scitranslmed.3002925

36. Kardani K, Bolhassani A, Shahbazi S. Prime-boost vaccine strategy against viral infections: Mechanisms and benefits. *Vaccine*. 2016; 34(4): 413-423. doi: 10.1016/j.vaccine.2015.11.062

37. Milligan ID, Gibani MM, Sewell R, et al. Safety and immunogenicity of novel adenovirus type 26- and modified vaccinia ankara-vectored Ebola vaccines: A randomized clinical trial. *JAMA*. 2016; 315(15): 1610-1623. doi: 10.1001/jama.2016.4218

38. Tapia MD, Sow SO, Lyke KE, et al. Use of ChAd3-EBO-Z Ebola virus vaccine in Malian and US adults, and boosting of Malian adults with MVA-BN-Filo: A phase 1, single-blind, randomised trial, a phase 1b, open-label and double-blind, dose-escalation trial, and a nested, randomised, double-blind, placebo-controlled trial. *Lancet Infect Dis*. 2016; 16(1): 31-42. doi: 10.1016/S1473-3099(15)00362-X

39. De Santis O, Audran R, Pothin E, et al. Safety and immunogenicity of a chimpanzee adenovirus-vectored Ebola vaccine in healthy adults: A randomised, double-blind, placebo-controlled, dose-finding, phase 1/2a study. *Lancet Infect Dis*. 16(3): 311-320. doi: 10.1016/S1473-3099(15)00486-7

40. Ledgerwood JE, DeZure AD, Stanley DA, et al. Chimpanzee adenovirus vector Ebola vaccine — Preliminary report. *N Engl J Med*. 2017; 376: 928-938. doi: 10.1056/NEJMoa1410863

41. Zhang Q, Seto D. Chimpanzee adenovirus vector ebola vaccine — Preliminary report. *N Engl J Med*. 2015; 373(8): 775-776. doi: 10.1056/NEJMc1505499#SA1

42. Group TPIW. A Randomized, controlled trial of ZMapp for Ebola virus infection. *N Engl J Med*. 2016; 375(15): 1448-1456. doi: 10.1056/NEJMoa1604330

43. Zhou Y, Sullivan NJ. Immunology and evolvement of the adenovirus prime, MVA boost Ebola virus vaccine. *Curr Opin Immunol*. 2015; 35: 131-136. doi: 10.1016/j.coi.2015.06.006

44. Ewer K, Rampling T, Venkatraman N, et al. A monovalent chimpanzee adenovirus Ebola vaccine boosted with MVA. *N Engl J Med*. 2016; 374(17): 1635-1646. doi: 10.1056/NEJMoa1411627

45. Geisbert TW, Geisbert JB, Leung A, et al. Single-injection vaccine protects nonhuman primates against infection with marburg virus and three species of Ebola virus. *J Virol*. 2009; 83(14): 7296-7304. doi: 10.1128/JVI.00561-09

46. Geisbert TW, Feldmann H. Recombinant vesicular stomatitis virus–based vaccines against Ebola and marburg virus infections. *J Infect Dis*. 2011; 204(Suppl 3): S1075-S1081. doi: 10.1093/infdis/jir349

47. Mire CE, Miller AD, Carville A, et al. Recombinant vesicular stomatitis virus vaccine vectors expressing filovirus glycoproteins lack neurovirulence in nonhuman primates. *PLoS Negl Trop Dis*. 2012; 6(3): e1567. doi: 10.1371/journal.pntd.0001567

48. Jones SM, Feldmann H, Stroher U, et al. Live attenuated recombinant vaccine protects nonhuman primates against Ebola and Marburg viruses. *Nat Med*. 2005; 11(7): 786-790. doi: 10.1038/nm1258

49. Wong G, Audet J, Fernando L, et al. Immunization with vesicular stomatitis virus vaccine expressing the Ebola glycoprotein provides sustained long-term protection in rodents. *Vaccine*. 2014; 32(43): 5722-5729. doi: 10.1016/j.vaccine.2014.08.028

50. Lai L, Davey R, Beck A, et al. Emergency postexposure vaccination with vesicular stomatitis virus–vectored Ebola vaccine after needlestick. *JAMA*. 2015; 313(12): 1249-1255. doi: 10.1001/jama.2015.1995

51. Agnandji ST, Huttner A, Zinser ME, et al. Phase 1 trials of rVSV Ebola vaccine in Africa and Europe. *N Engl J Med*. 2016; 374(17): 1647-1660. doi: 10.1056/NEJMoa1502924

52. Skrip LA, Galvani AP. Next steps for Ebola vaccination: Deployment in non-epidemic, high-risk settings. *PLoS Negl Trop Dis*. 2016; 10(8): e0004802. doi: 10.1371/journal.pntd.0004802

53. Regules JA, Beigel JH, Paolino KM, et al. A recombinant vesicular stomatitis virus Ebola vaccine - Preliminary report. *N Engl J Med*. 2015; 376: 330-341. doi: 10.1056/NEJMoa1414216

54. Huttner A, Dayer JA, Yerly S, et al. The effect of dose on the safety and immunogenicity of the VSV Ebola candidate vaccine: A randomised double-blind, placebo-controlled phase 1/2 trial. *Lancet Infect Dis*. 2015; 15(10): 1156-1166. doi: 10.1016/S1473-3099(15)00154-1

55. Henao-Restrepo AM, Longini IM, Egger M, et al. Efficacy and effectiveness of an rVSV-vectored vaccine expressing Ebola surface glycoprotein: Interim results from the Guinea ring vaccination cluster-randomised trial. *Lancet*. 2015; 386(9996): 857-866. doi: 10.1016/S0140-6736(15)61117-5

56. Li H, Ying T, Yu F, Lu L, Jiang S. Development of therapeutics for treatment of Ebola virus infection. *Microbes Infect*. 2015; 17(2): 109-117. doi: 10.1016/j.micinf.2014.11.012