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Is Prime Boost Strategy a Promising Approach in HIV Vaccine Development?

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Abstract

Since the discovery of HIV three decades back, the quest for HIV vaccines has remained unquenched. There has been a transition of preferred approaches from candidates capable of inducing neutralizing antibody (Nab) or cytolytic T cell (CTL) response to vaccines that can induce broad spectrum responses. Heterologous prime boost strategy is believed to induce broad spectrum immunity of higher magnitude and breadth to effectively counter HIV diversity and hence is being studied extensively in the HIV vaccine field. It is important to understand factors affecting the immune responses generated by the prime-boost regimens to get leads for developing effective regimens. This review focuses on the results of completed clinical trials based on the three most frequently used prime-boost regimens, vector (ALVAC)/protein, DNA/vector (MVA) and DNA/vector (Ad5). It will also discuss probable protective immunological responses responsible for efficacy of the vaccine and role of prime boost strategy in eliciting them.

Keywords: CTL response; Envelope; Heterologous HIV vaccine; Neutralizing antibody; Prime-boost Strategy; Protein subunit; Vector

Introduction

HIV prevention strategies to reduce the spread of HIV remain a global public health priority. An effective HIV vaccine, if available, would have several advantages over behavioral and other biological prevention strategies as it would not need sustained behavior changes as well as strict adherence for the efficacy, in addition to probably providing long term protection. Although inactivated and live attenuated vaccines are being effectively used for other viral infections, they are not being considered favorably in case of HIV because of safety concerns. Hence most of the efforts towards HIV vaccine development have been focused on newer strategies such as synthetic envelope protein subunits or recombinant viral vectors carrying HIV-specific inserts or naked DNA [1]. Although most of these vaccine candidates have failed to elicit effective immune responses when used alone [2,3], their combined use has been shown to strengthen and broaden HIV-specific immune responses [4-6]. Such a combination strategy is known as prime-boost strategy where the immune system primed by one vaccine candidate is boosted with either the same (homologous) or a different (heterologous) vaccine candidate.

More than 100 non human primate and human clinical trials have been conducted so far to test the safety, immunogenicity and efficacy of different combinations of vaccine candidates [7]. The results of clinical trials have often been shown to differ from those seen in animal studies indicating critical need for reviewing them to understand the immune responses elicited by the vaccine candidates for use in humans [8]. Hence this review is focused on discussing the outcomes of different prime boost HIV vaccine clinical trials and factors responsible for them, which would have implications in guiding future vaccine trials based on prime boost strategy.

Basis of Prime Boost Strategy

Boosting of immune responses by vaccines results in generation of larger numbers of effector cells required for mediating protection against pathogens at the time of infection [9]. Homologous strategy effectively boosts the humoral immunity but fails to boost cellular immunity (CMI). While heterologous prime-boost approach is known to effectively boost CMI, especially when vector based vaccine candidates are used, as it minimizes the interference by anti-vector immunity generated after priming (illustrated in Figure 1). Apart from enhancing the effector cells quantitatively, qualitative differences in secondary memory cells are also seen after the boosting. Secondary memory CD8 T cells, in contrast to primary memory cells, traffic much more efficiently to peripheral tissues and exhibit enhanced cytolysis facilitating effective countering of pathogens at the site of entry [10]. Additionally, heterologous prime boost strategy results in synergistic enhancement of immune response resulting in an increased number of antigen-specific T cells, selective enrichment of high avidity T cells and increased breadth as well as depth of the immune response [11,12].

However, heterologous prime boost regimens are still at the stage of clinical research and no regimen has been adopted in the immunization programs until now, as the critical evidence of translation of all these advantages is lacking. One of the major limitations of the prime boost strategy is its complex design. Multiple factors can contribute to its efficacy, which include combination of vaccine candidates, order of their administration, vaccine dose, interval between various antigen exposures, route of vaccine administration, pre-existing immunity to the vectors and relatedness of epitopes between the prime and booster antigens. Other limitations of the strategy include the requirement of multiple dosages of different vectors which might add to potential side effects and could be challenging for a vaccination programme. A phenomenon of 'original antigenic sin' may also occur where by immune response to original antigens present in the prime interfere in

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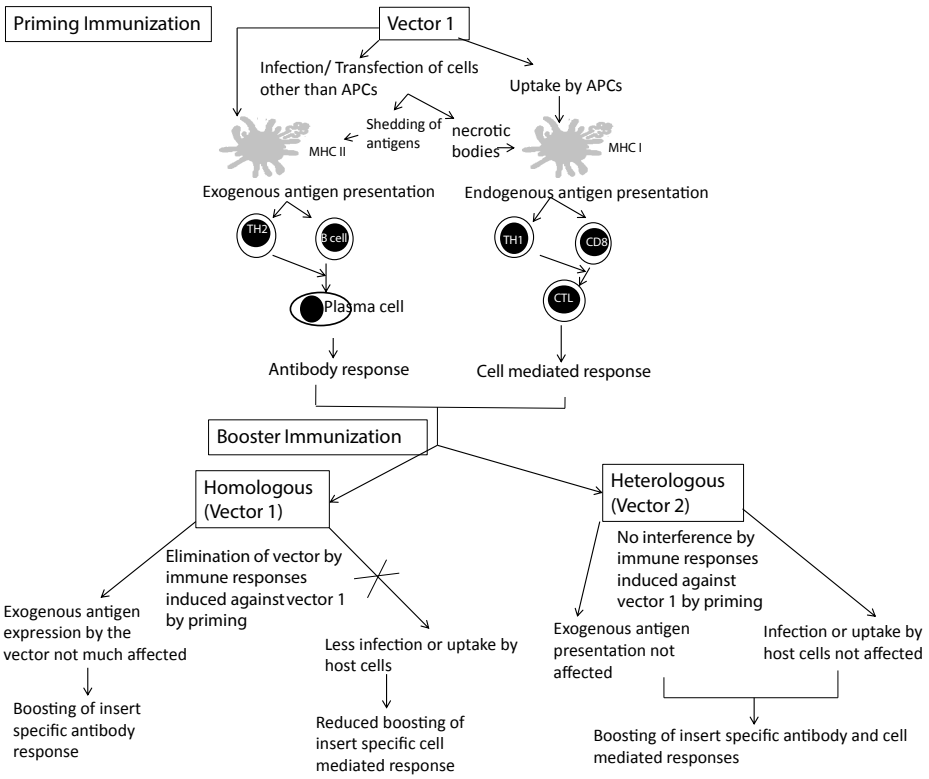

Figure 1: The role of heterologous prime boost strategy in overcoming anti-vector immunity
Vector based vaccine candidates infect host cells including antigen presenting cells. Induction of immune response depends on antigen presented by host cells after uptake of the vector. Cellular and humoral immune responses are induced against vector and insert specific antigens through class I and class II MHC molecules, respectively, upon the antigen presentation. After boosting of immune response with the same vector, uptake of the vector by host cells gets hampered because of elimination of the vector by anti-vector immunity. This further affects antigen presentation and boosting of insert specific immune responses by the vector. Conversely, if a different vector is used for boosting, anti vector immunity induced by the prime cannot eliminate an antigenically different vector. Hence, the uptake as well as antigen presentation by the host cells is not affected, leading to boosting of insert specific immune responses.

the response elicited to new antigens present in the boost, if different insert sequences are used for priming and boosting [13].

Clinical trials of HIV-1 candidate vaccines using prime boost strategy

HIV vaccine trials are primarily focused on generation of neutralizing antibodies to prevent the establishment of infection or generation of T-cell responses effective in reducing viral burden in the post-infection phase [14,15]. However, since both these strategies alone failed to show protection from HIV infection [2,3,16], their combination was thought to be beneficial for protection and hence was evaluated in clinical trials by combining different strategies. The most frequent combinations tested in clinical trials include vector prime protein boost and DNA prime vector (pox or adenovirus) boost combinations.

Vector prime protein boost

Clinical trials carried out using vector prime protein boost regimen have mainly focused on the pox virus vectors for priming. Pox virus vector constructs have the ability to induce CTLs in humans but they have not been shown to elicit high-titer neutralizing antibodies [4,17,18]. However, the use of an envelope protein boost after the pox vector prime has been reported to generate higher levels of binding and homologous neutralizing antibodies in addition to development of antibody-dependent cell mediated cytotoxicity (ADCC), and helper T cell responses in several phase I clinical trials [18-20]. Non-replicating poxvirus vectors, including Modified Vaccinia Ankara (MVA), and the genetically modified NYVAC vector, fowl pox and canary pox (ALVAC) vectors are preferred in clinical trials over replication competent vaccinia vector because of its safety concerns and poor immunogenicity, possibly due to existing anti-vector immunity resulting from global smallpox vaccinations [21,22]. Among all pox viruses, several Canarypox vector based constructs with multiple HIV-1 gene inserts have undergone extensive safety and immunogenicity studies in humans [23,24]. Although the Canarypox prime-protein boost regimens have been shown to induce increased frequency and magnitude of HIV-1 lympho-proliferative activity, neutralizing activity and ADCC, the CTL responses induced by them were limited. Hence, several modifications have been made in the construct to improve CTL responses as illustrated in Figure 2. A phase III trial, RV144, using Canarypox (vCP1521) prime and AIDSVAX B/E boost has demonstrated modest protective efficacy when tested in Thailand [25]. But this strategy failed to control viremia or CD4 cell loss after the breakthrough infections among those who received the vaccines indicating inability of this strategy to alter the course of the disease. The protection in RV144 trial appeared to be short lasting, [26] and it would be interesting to evaluate the effect of additional boosters in RV144 trial participants on recall responses and continuing protection among them. Indeed, in the AVEG studies (AIDS Vaccine Evaluation Group), a recall antibody response after delayed rgp160 boost at 4-5 years of ALVAC vaccinations has indicated induction of long term memory B

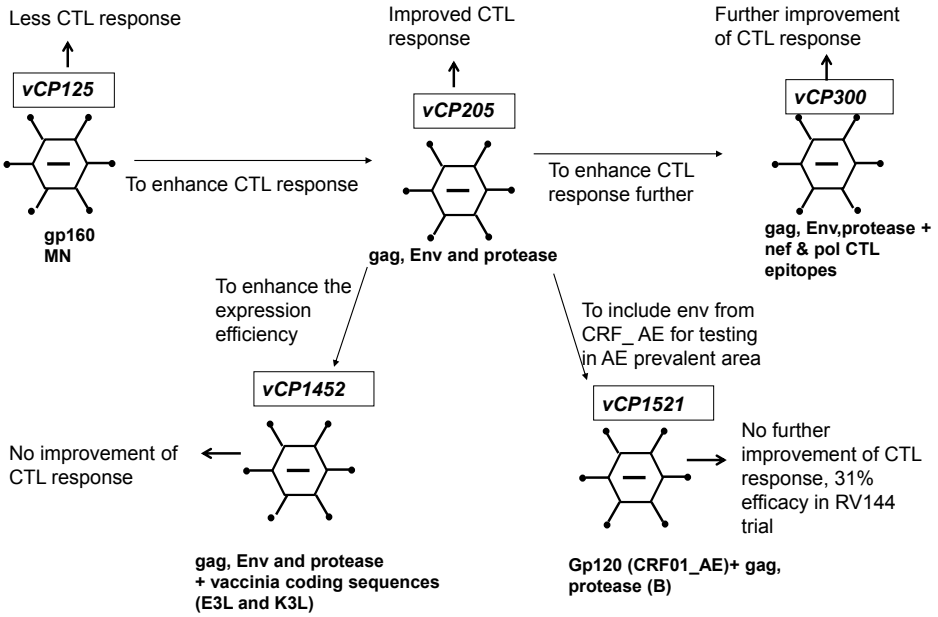

Figure 2: The ALVAC–HIV constructs evaluated in combination with protein boost in clinical trials
Different ALVAC construct developed and used as a prime in association with protein boost are shown in the figure. Different HIV genes inserted in these constructs are mentioned at the bottom of each construct.

cell responses [27].

DNA prime and & Poxvirus vector boost strategy

DNA prime-viral vector boost regimens have become the primary choice for inducing T cell based immune responses [11,28,29]. Among all Poxvirus vector based vaccines, MVA based vaccines have been evaluated in combination with DNA priming in multiple clinical trials. However, the MVA vectors used in different trials differed from one another in terms of passage numbers, insertion sites, type of promoter used and the inserted HIV genes leading to non-comparability of the results reported in different clinical trials [30]. Advantage of adding the DNA prime to MVA based regimen remains questionable as it has been shown to induce only marginally higher T cell responses as compared to the homologous MVA boosting strategy [31,32]. Also antibody responses induced by the DNA/ MVA strategy were found to be inferior as compared to those induced by the homologous MVA boosting strategy [31,32]. Among the other poxvirus based vaccines evaluated in clinical trials, NYVAC based regimens appear to be promising in eliciting the immune responses [33-35]. On the other hand, fowl pox based regimens have been shown to be poorly immunogenic in humans [36,37] while they induced effective CD4 and CD8 T-cell responses in animal models [38].

Strategies using DNA and Adenoviruses

Among the Adenoviral vector vaccine candidates, replication-defective Ad5 candidate developed by the Merck group has been studied most extensively in human trials. This construct demonstrated good immunogenicity in Phase I clinical trials and reduced viral load in the SHIV/NHP model [13,39-42]. However, it failed to prevent new infections as well as to reduce post-infection viral RNA levels in the vaccinated individuals in phase IIb, test-of-concept, STEP trial [16]. In addition, participants with pre-existing antibodies against Ad5 vector showed increased HIV infection rates in the study, the cause of which is being evaluated at present. The pre-existing immune responses might have played a role in this as they are known to interfere in the HIV specific immune responses induced by Ad5 vector [41]. High rates of pre-existing humoral immunity (as high as 85% in South Africa) to Ad5 have been shown in many parts of the world [43]. The heterologous prime boost strategy using DNA prime and Ad5 boost was thought to circumvent the problem of pre-existing immunity. It has been shown that the pre-existing Ad5 neutralizing antibodies did not affect the frequency and magnitude of T cell responses in the DNA/rAd5 prime-boost recipients, as compared to participants who received rAd5 alone [44]. Although there are conflicting reports regarding the increased magnitude of immune response by DNA/Ad5 strategy over Ad5 vaccine alone, the strategy was successful in inducing both CD4 and CD8 responses contrary to the DNA and Ad5 vaccine candidates alone which generated only CD4 or CD8 responses respectively [45,13]. Since the broad spectrum immune response consisting of both CD4 and CD8 responses is desirable for protection against HIV, this strategy has been considered to be more suitable than homologous Ad5 vaccinations. However, in spite of robust immune responses induced by DNA/Ad5 strategy in phase I and phase II trials [46], the strategy failed to show protection from new infections in a phase IIb, HVTN 505 trial, which had to be terminated prematurely [47]. Unfortunately the HVTN 505 trial, like the STEP trial, showed a statistically insignificant trend towards more infections among the vaccine recipients [48].

Other adenovirus serotypes like Ad26 and 35, less commonly associated with human disease, are also being explored as vaccine candidates to obviate the interference from pre-existing immunity against Ad5 [49]. However, Nabs to Ad5 have also been shown to hamper $CD4^+$ T-cell responses to DNA/rAd35 combination [50].

Factors affecting immunogenicity endpoints of the prime boost regimens

The clinical trials described in the preceding paragraphs differed

from each other in terms of vaccine candidates used, doses, schedule, route and mode of vaccination as well as the population in which the trials were conducted. Understanding these factors would help in optimizing the vaccination schedule to obtain high and persistent immune responses.

Vaccine dose

Dose of a prime candidate may not be important as it was not found to influence the final immunogenicity results in the trials with all the three types of regimens [17,45,51]. This could be possible because higher antigen doses at priming generally favor the induction of effector cells, whereas lower doses may preferentially drive the induction of immune memory [52]. Hence higher dose of a prime, although desirable for immediate responses, may affect development of memory cells and adversely hamper the effect of high dose. Contrary to the prime dose, higher dose of the booster has been shown to induce higher magnitude of immune response in ALVAC/protein and DNA/MVA trials [51,53] as the greater availability of antigen might be driving higher number of memory B cells into differentiation, thereby amplifying the response. However, this effect was not seen when DNA/AD5 strategies with Ad5 doses of 10^{10} and 10^{11} were compared [46]. This could be because of development of immune tolerance at such high doses. Similar results were also observed when only Ad5 based strategy was evaluated in a clinical trial [54].

Immunization schedule

It was observed that late boosts at 5 and 6 month interval induced higher T cell response as compared to the early boosts given at 2 and 3 months interval in one of the trials with DNA/MVA prime/boost regimen [55]. The delayed boosting is helpful in avoiding interference in the primary responses induced by the prime [52]. It has been observed that although closely spaced (1–2 weeks) primary vaccine doses cause a rapid induction of immune response, the response is less persistent than when the same numbers of vaccine doses were given at longer intervals (1–2 months) [52]. A minimal interval of 4–6 months may also ensure optimal affinity maturation of memory B cells [52]. One of the DNA/Ad5 trials showed that the boosters, as late as at 35 and 94 weeks, also increased the frequency and magnitude of T cell and antibody response [45], which was, in fact, better than in the other trials which employed boosting at 24 weeks.

The number of doses required for inducing optimal immune response may differ based on the vaccine candidate. It has been found that 4 doses of ALVAC and 2 doses of DNA were optimum for inducing CTL and CD4 helper T cell responses respectively when used for priming [56,57]. There are conflicting reports regarding the number of MVA doses required for boosting. One study reported higher T cell response rate after 2 MVA doses [56], whereas diminution of response rate and magnitude after second MVA dose was reported in another trial [58]. Number and timing of protein boosts differed considerably in ALVAC/protein based trials as multiple combinations were evaluated with either sequential or simultaneous boosting. Although the simultaneous boosting showed early antibody responses [59], sequential boosting schedules were reported to elicit higher magnitude of neutralizing antibody response [57].

Route of administration and delivery systems used

Different routes and delivery systems have been evaluated for DNA constructs in prime–boost strategy. It has been shown that DNA constructs, administered by intra-dermal route induced better immune response compared to the intramuscular or subcutaneous routes [60,61]. Immunogenicity of DNA constructs depends on processing and presentation of antigens by antigen presenting cells (APCs) and the skin, unlike muscle tissue, has a large population of resident antigen presenting cells (APCs) that can facilitate the induction of vaccine-specific immune responses [62,63]. Biojector and electroporation for delivering DNA have also shown to enhance antigen presentation by targeting larger area and enhancing uptake by the cells by the transient formation of pores in the cell membrane, respectively [62,64]. The biojector delivery system was observed to be better than administration by needle/ syringe as well as by electroporation in two HIV clinical trials [58,65]. A combination of biojector and electroporation has been shown to overcome dose restriction of DNA vaccines in preclinical studies whereby immune responses were shown to be enhanced when the dose of DNA was increased in clinical trials [66].

Adjuvants

Adjuvants are components of vaccines used for potentiating and/or modulating the immune responses to an antigen. Since DNA vaccines are weakly immunogenic, different adjuvants like Al_3PO_4 or CRL1005 and immuno-modulators like GM-CSF were evaluated for enhancing their immunogenicity in the prime boost trials without success [13,51]. These adjuvants had shown promising effects in animal studies [51]. This also highlights the importance of determining immune responses in clinical settings. One of the trials which evaluated two adjuvants namely MF59 and Alum for the protein boosting showed that MF59 adjuvanted protein subunit candidate induced better response than using Alum as an adjuvant [53,67]. MF59 has also shown to be more potent than alum based adjuvants in inducing both antibody and T-cell responses when evaluated as an adjuvant for flu vaccine and is being currently used in flu vaccine preparations [68].

Pre-existing immunity against the vectors

Prime boost strategy has been thought as one of the ways to circumvent the pre-existing immunity to vectors. However, DNA/Ad5 based clinical trials have provided inconclusive evidence in this regard [44,13]. For ALVAC/protein based strategy, one study reported no significant differences in frequency or level of immune responses to ALVAC (with or without protein boost) between Vaccinia-naive or Vaccinia immune individuals [17], while another study reported decreased magnitude of Nabs in Vaccinia immune individuals compared to Vaccinia-naive [57]. Lower magnitude of cellular immune responses in persons with a history of Vaccinia vaccination has been reported in one of the DNA/MVA trials, which has been thought to be contributed additionally by effect of age in these individuals [51] indicating multifactorial etiology responsible for such variations.

Relatedness of insert sequences or heterologous inserts

Not enough clinical data are available for comparing immune responses induced by heterologous insert sequences. The results of the ALVAC/protein based clinical trials having insert gp120 sequences from the same or different strains of HIV-1 do not differ much from each other. Although they are thought to improve global epitope coverage or cellular immune breadth, they failed to demonstrate this in an animal study [69]. The phenomenon of original antigenic sin also needs to be kept in mind before devising strategies based on heterologous inserts.

Can Prime Boost Vaccine Strategy Fulfil the Possible Criteria for an Ideal HIV Vaccine to be Protective?

Although immunogenicity results are available from multiple phase I and II trials based on the prime boost strategy, they are not sufficient

to predict the efficacy of the strategy. Analyses of immune responses observed in large scale efficacy trials are, therefore, important as they are likely to provide clues about protective immune responses in HIV infection. The difference in the immune responses elicited in RV144 prime boost trial that demonstrated modest protection and VAX003/VAX004, AIDSVAX alone, trials need to be carefully analyzed to delineate the factors that might have contributed to the protection. Based on results of such efficacy trials as well as data from animal studies using SIV challenge, different factors that possibly play role in efficacy of the immune response can be deciphered.

Type of immune response

Although broadly neutralizing antibodies are considered to be an important component of vaccine induced immune responses, it has not been possible to induce them in any of the clinical vaccine trials conducted so far. Antibody based phase III vaccine trials have utilized monomeric gp120 presenting linear antigens, not considered to be optimum for induction of neutralizing antibodies. Since conformational epitopes are considered to be more potent in inducing neutralizing antibodies, candidates presenting envelope antigens in their native configuration would be more appropriate for vaccine development. Virus-like particles and trimeric gp140 antigens have shown promise in induction of neutralizing antibodies [70,71] and can be employed in prime boost combinations for effectively inducing broadly neutralizing antibodies.

Non-neutralizing antibodies have also been shown to play a role in protection from HIV infection in the RV144 trial. IgG antibodies specific to the V1/V2 region of HIV-1 gp120 correlated with a decreased risk of infection with evidence of a virus sieve effect in infected vaccine recipients at this gp120 region [72,73]. Although the exact mechanism mediating protection by these antibodies in the trial is not yet known, they are thought to block T-cell associated integrin, a4b7, which is involved in HIV-1 entry in activated CD4+ T-cells [74,75]. The role of these antibodies in mediating protection from HIV needs to be confirmed further.

As against the neutralizing antibodies, responses like CTLs and ADCC act after infection of the cells and eliminate the infected cells decreasing overall burden of HIV by blocking further multiplication of virus. RV144 trial data showed that high level of ADCC activity was inversely correlated with infection risk. For mediating such activity, specificity and Fc related functions of the antibodies would be the important determinants. Antigens expressed on cells would be important for mediating ADCC activity and it may be possible that these antigens were expressed by infected cells upon uptake or infection by ALVAC vector and antibodies against them were subsequently boosted by gp120 protein boost. It was also observed that the IgG isotypes in case of RV144 trial participants were IgG1 and IgG3, which have the ability to bind and stimulate the NK cells efficiently through binding with CD16 [76]. Compared to the RV144 results, VAX003 trial participants had higher titres of IgG4, which show poor ADCC activity [76]. The future vaccine trials based on vector prime protein boost strategy should be equipped to explore the role of ADCC activity in the protection.

Like ADCC, CTLs are also important in eliminating HIV infected cells and their role in controlling HIV progression has been well documented from studies on Long Term Non Progressors (LTNPs), exposed but uninfected individuals and in non human primate models [77-79]. However, it has not been possible to attribute a role to CTLs in controlling HIV infection in clinical vaccine trials conducted so far. In theRV144 trial showing moderate protection, a CTL response was reported in only 19.7% of vaccinees [25]. On the other hand, the Ad5 based STEP and HVTN 505 trials failed to prevent HIV-1 infection or reduce early viral level, in spite of induction of a CTL response in 75% and 64% of the vaccinees respectively [16,47]. However, despite the lack of vaccine efficacy in the STEP trial, participants with specific human leukocyte antigen (HLA) alleles demonstrated an evidence of vaccine-elicited immune pressure on the founder virus resulting in specific escape mutations. They also had lower viral load highlighting the role of CTL response in controlling viral multiplication in the trial [80].

Dynamics of immune response

Antibodies are usually long-lived and hence once generated are usually available at the time of exposure to deal directly with the incoming viral inocula resulting in protection [81]. However, this is not the case with CTL based vaccines. Effectors and effector memory cells (TEM), which are considered to be important for immediate action against any pathogen are short lived and tend to disappear after the clearance of the antigens leaving behind central memory T cells (TCM). Hence soon after vaccination with the non persistent vaccine candidates, only TCM type of cells persist, which shows inherent delay in development of anti-viral effectors limiting the utility of CTL based vaccines [81]. On the contrary, persistent vectors like Rhesus cytomegalovirus (RhCMV) were shown to induce persistent, high-frequency, SIV-specific TEM responses at mucosal sites resulting in stringent control of highly pathogenic SIV_{MAC239} infection after mucosal challenge in rhesus macaques when used alone or in prime boost combination [82,83]. Hence the use of persistent vectors needs to be evaluated further in clinical trials for developing of effective CTL based vaccines. However, such vaccine approaches also carry a risk of developing CD4 TEM providing a higher frequency of activated target cells at mucosal sites [81]. Hence vaccine regimens eliciting a predominant CD8 response would be important for balancing potentially infection-suppressing and infection-facilitating mechanisms.

Site of immune response

Mucosal immune responses are considered to be important for restricting the virus multiplication at the site of entry before systemic dissemination occurs. The factors which favor development of mucosal immune responses include the mucosal or trans cutaneous immunization and the replicating nature of the vaccine agents [84,85]. A prime boost strategy with heterologous routes of administration based on the combination of mucosal and parenteral delivery has been attempted in a murine model for inducing immune responses at both mucosal and systemic levels [86]. Although mucosal route for vaccination is desired for its ease of administration and development of local immunity, mucosal vaccinations are faced with safety concerns and problems of lesser efficacy [87]. Therefore, only a few vaccines have become available for mucosal use until now. Since many of the HIV vaccine trials have used live viral vectors and percutaneous route, mucosal immune responses might have been generated in the vaccinees. Unfortunately information on mucosal immune responses in HIV vaccine clinical trials is scarce. It is important that the future trials should assess immune responses generated at the mucosal level.

Prime boost strategy has also been shown to generate secondary memory cells which tend to localize in peripheral tissues causing effective an immune response at the site of infection [10]. However, these cells tend to localize poorly in lymph nodes which are the major sites of HIV replication [10]. Hence these cells may not play a role in controlling viremia once the virus escapes the mucosal immune

responses. Hence it would be important to balance mucosal as well as systemic T cell responses in order to effectively control the infection. Other limitations of mucosal immune responses include generation of activated CD4 cells at mucosal sites, which might serve as potential targets for HIV infection. Also mucosal reactions can increase blood-borne IgA secreting plasma cells which are known to be derived from mucosal immune responses [88]. HIV-1 Env-specific plasma IgA responses have shown direct correlation with HIV infection risk in the RV144 trial, which could be by blocking protective IgG antibodies mediating ADCC activity [89].

Magnitude of immune response

Potent immune responses of higher magnitude are desirable to deal with a higher inoculum of the virus. Prime-boost combination vaccines have been found to elicit a dramatic enhancement in the magnitude of anti-viral $CD8^+$ T cell responses after infection (often >10-fold) [81]. Hence a prime boost strategy can be used for enhancing potency of the cell mediated immune responses.

Breadth of immune response

The vaccine strategies that expand breadth of cellular immune response have been considered to be critical for achieving immunologic coverage of the enormous global genetic diversity of HIV-1 [90]. Importance of breadth of immune response has also been highlighted from studies in chronically infected patients and macaque models [91,92]. The STEP trial, which failed to show protection, demonstrated limited breadth of response [93]. To increase the breadth of response and to cover global HIV-1 sequence diversity, polyvalent 'mosaic' antigens have been designed, which have been shown to induce markedly augmented breadth and depth of response without compromising the magnitude of T cell responses in Rhesus monkeys [90]. Protective efficacy of the mosaic antigens has also been demonstrated in Macaques [94]. Clinical studies evaluating Ad26/MVA regimens expressing HIV -1 mosaic antigens have been planned and would provide clinical data in this regard.

Immune responses against conserved epitopes

Targeting conserved epitopes by the immune response has also been considered to be an important component of vaccine induced immunity to overcome HIV diversity and mutations. However, conserved epitopes have been found to elicit subdominant responses during both primary and chronic infection [95] as well as in a vaccine trial. The immune response has been seen to be biased towards non conserved epitopes, which might mask responses to conserved epitopes enabling HIV to escape immune surveillance mechanisms [96]. The prime boost strategy could be exploited to boost specifically the responses against the conserved epitopes by using these epitopes in the boost candidates.

Analysis of correlate of risk

As against the correlates of protection, analysis of correlates of increased risk of HIV acquisition has become imperative as a result of findings in Ad5 based STEP and HVTN 505 trials. The STEP study reported an increased risk of HIV acquisition among MSM who were uncircumcised or had neutralizing antibodies to Ad5 at the enrolment [16]. In spite of extensive research to identify biological reasons for such an increased risk, no evidence is available yet. The subgroups of participants showing enhanced risk in STEP trial were excluded from HVTN 505 trial. However, a trend of increased risk of HIV acquisition was also reported in the HVTN 505 trial demonstrating the susceptibility of these subgroups for the enhanced risk. Based on the HVTN 505 results, the results of the STEP trial also need to be re-evaluated with a different focus as the pre-existing immunity against Ad5 alone may not be playing a role in enhancing the risk of HIV acquisition. Possibility of occurrence of such a risk with other adenovirus serotypes should also be ruled out before proceeding to the clinical trials using these constructs.

Conclusion

The available evidence suggests that the heterologous prime-boost vaccination approach has some promise in HIV vaccine development. However, the prime-boost strategy has major operational and analytical complexities. Also the efficacy results have churned out surprises quite in contrast to the preclinical as well as immunogenicity data available from phase I and II trials. This may be due to the lack of confirmed knowledge about the correlates of protection in HIV infection. Hence, analysis of correlates of protection is critical for designing effective vaccine trial strategies. Two vaccine efficacy trials based on prime boost strategy, the RV144 and HVTN 505 trials, have demonstrated exactly opposite outcomes. The results of these trials need to be evaluated carefully to determine correlates of protection and increased risk of HIV acquisition for balancing protective and enhancing mechanisms in future vaccine trials. Up till now, only RV144 vaccine trial has been successful in demonstrating moderate, but short lasting protection against HIV infection. The search for the most appropriate regimen for eliciting effective and sustained immune responses must be continued till an effective preventive vaccine strategy is devised.

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