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REVIEW

Challenges in the search for an HIV vaccine

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Abstract. Considerable progress has been made over the past several years in the development of an HIV vaccine. As a result, a growing number of vaccine modalities are being investigated in pre-clinical and phase I/II clinical trials. However, a number of major scientific challenges still remain. It is widely believed that the ideal vaccine should elicit both neutralizing antibodies and cytotoxic T lymphocytes (CTL) against diverse isolates of HIV, but the precise correlates of immunity have not been defined. Recombinant live vector-based vaccines and plasmid DNA vaccines have been shown to induce CTL, either alone or in combination, and these CTL-based vaccines have shown partial protective efficacy in nonhuman primates challenge studies. An immunogen that elicits broadly reactive neutralizing antibodies, however, has yet to be developed.

Key words: HIV, Immune response, Vaccine

Abbreviations: AIDS = acquired immunodeficiency disease syndrome; CTL = cytotoxic T lymphocytes; HIV = human immunodeficiency virus

Introduction

The development of a safe and effective human immunodeficiency virus (HIV) vaccine has proven a major scientific challenge. Given the rapidly expanding HIV pandemic, the need for a prophylactic HIV vaccine is paramount, particularly in the developing world. In the Western world, antiretroviral therapy is responsible for major declines in AIDS-related morbidity and mortality. However, despite recent advances in bringing antiretroviral therapy to resource-poor settings, these drugs are still not available to the vast majority of individuals in these areas where they are needed most.

The most effective way to control the spread of the HIV epidemic will be the development of a prophylactic vaccine. It is generally agreed that an effective vaccine will likely need to elicit both humoral and cellular immune responses. Neutralizing antibodies bind free virus particles and eliminate them through various effector mechanisms. CD8+ cytotoxic T lymphocytes (CTL) kill HIV-infected cells, and CD4+ helper T lymphocytes serve a critical role in orchestrating the immune response. The precise immune correlates of protection, however, have not yet been defined.

The ultimate goal of an HIV vaccine is to achieve sterilizing immunity. However, this level of protection is not even achieved with most clinically licensed vaccines. A more realistic goal may be to develop a vaccine that lowers viral loads and prevents clinical disease progression. Recent clinical and pre-clinical studies have demonstrated that virus-specific adaptive immune responses are critical for immune control of viral replication. For example, people who are highly exposed to HIV but remain uninfected have been reported to have HIV-specific cellular immune responses [1, 2]. Moreover, potent CTL responses have been observed in long-term non-progressors [3]. Studies in non-human primates have shown that passively transferred neutralizing antibodies can provide complete protection against infection [4–7]. Vaccine-induced cellular immune responses have been shown to suppress viral replication but not provide sterilizing immunity against pathogenic viral challenges [8–11].

HIV neutralizing antibodies

HIV-specific neutralizing antibodies likely play a role in controlling viral replication, but the development of immunogens capable of eliciting broadly reactive neutralizing antibodies has proven extraordinarily difficult. HIV-infected patients generate antibodies to a variety of HIV proteins following infection. However, these antibodies are often not neutralizing and are often directed against viral debris [12]. Neutralizing antibodies to HIV are mainly directed against the envelope proteins, gp120 and gp41. These antibodies often recognize epitopes in the third variable region (V3 loop) or the CD4 binding domain, but antibodies directed against the V3 loop more efficiently neutralize laboratory strains of HIV-1 than

primary HIV isolates [13]. Moreover, it has been shown that HIV is very efficient at escaping neutralizing antibodies by accumulating mutations in the envelope gene. Shielding of the conserved regions of the envelope protein by the other two variable loops (V1 and V2 loop) [14] and by extensive glycosylation [15] limits the accessibility of neutralizing antibodies. Interestingly, modification of glycosylation patterns has been reported as a novel neutralization escape mechanism [16].

Overall, the ability of HIV to escape neutralizing antibody responses and to shield conserved epitopes makes it difficult to develop a vaccine that is able to elicit neutralizing antibodies. Moreover, the genetic variability of HIV-1 is an enormous challenge that needs to be overcome. At present, no vaccine has been developed that reliably elicits neutralizing antibodies that recognize a broad diversity of primary isolates. Whether it is possible to elicit such broadly reactive neutralizing antibodies by vaccines is not known, although a wide variety of approaches are currently being explored in pre-clinical studies.

HIV CTL responses

Accumulating evidence has confirmed the importance of virus-specific CTL in controlling HIV replication in humans and SIV replication in rhesus monkeys. In particular, it has been convincibly shown that CTL are critical in controlling primary viremia in both human and monkeys [17, 18]. High levels of HIV-specific CTL have also been observed in long-term non-progressors [19]. These data suggest that vaccine strategies should elicit potent HIV-specific CTL responses. Recent non-human primate challenge studies have suggested that CTL-based vaccines will likely not protect against HIV infection, but they may have the ability to control viral replication and slow clinical disease progression [8–10].

It is possible that these CTL-based vaccines may slow disease progression and reduce the rate of HIV transmission [20]. However, these vaccines may be limited by a lack of durability of immune control. For example, HIV might eventually escape dominant CTL responses by viral mutations within CTL epitopes [21–23]. In fact, it has already been shown that viral escape from CTL can result in eventual AIDS vaccine failure in the rhesus monkey model [11]. Viral escape from CTL has also been shown to result in eventual progression towards AIDS in humans [22, 23].

The loss of HIV-specific CD4+ T cells in HIV-1 infected patients may also contribute to the eventual failure of CTL-based vaccines. High frequency CD4+ T lymphocyte responses are typically observed in long-term non-progressors [24]. In contrast the majority of HIV patients have poor CD4+ T lymphocyte responses [25]. These data suggest that HIV-specific CD4+ T helper cells are essential for the functionality and survival of CTL, and that maintaining these cells will be critical for vaccine-mediated immune control of HIV-1.

Vaccine modalities

A large number of vaccine modalities are currently being explored in both pre-clinical and clinical trials. Recombinant envelope proteins have been the most extensively studied candidate HIV vaccines. It was hoped that these vaccines would elicit neutralizing antibodies that would be able to prevent infection. However, these envelope proteins have different conformational structures than the native HIV envelope trimers on the surface of virions. The challenge of developing effective gp120-based vaccines has recently been highlighted by the failure of recombinant gp120 in two recent phase III human trials [26]. This was not surprising, however, since recombinant gp120 does not elicit broadly reactive neutralizing antibodies or virus-specific CTL responses.

Recombinant live vectors and plasmid DNA vaccines are currently being developed to elicit virus-specific CTL responses. CTL responses are frequently detected against internal proteins such as gag and pol and regulatory proteins such as nef in long-term non-progressors [27–29], suggesting that these genes should be included in an HIV vaccine. Attenuated poxvirus vectors have been developed that have the capacity to insert multiple HIV genes such as env, gag, pol and rev [30, 31]. MVA, NYVAC and fowlpox vectors have been shown to elicit CTL responses, either alone [32] or in combination with DNA vaccines [9, 33]. The canarypox vector ALVAC has been shown to induce low frequency but detectable HIV-specific CTLs in phase I/II clinical trials [34–36], but the immunogenicity of this vector was not potent enough to proceed with phase III clinical testing as a single-modality vaccine.

Recombinant adenovirus vectors have also been demonstrated to elicit high frequency CTL responses in non-human primates and phase I clinical trials [10, 37]. A major limitation of this approach, however, is that pre-existing immunity to the adenovirus serotype 5 vector will likely suppress the immunogenicity of these vaccines [38]. To circumvent this problem, non-human adenoviral vectors [39], and adenoviral vectors from rare human serotypes are being developed [40, 41].

Other recombinant live vectors are also being developed, including Semliki Forest virus [42], Venezuelan equine encephalitis [43] and adeno-associated virus. DNA vaccines have also been developed as candidate HIV vaccines. Although these DNA vaccines are not very immunogenic on their own, they have proven to be effective when used as a prime in

combination of viral vectors [9, 33, 38] or when adjuvants are added [8, 38].

Clinical trials are currently evaluating several recombinant live viral vector-based vaccines and plasmid DNA vaccines. These include phase I/II trials of a DNA prime/ MVA boost vaccine in the UK and Kenya sponsored by International AIDS Vaccine Initiative (IAVI). A recombinant Ad5 vaccine produced by Merck is currently in phase I clinical trials in the US**.** The Vaccine Research Center (VRC) at the National Institutes of Health (NIH), has two phase I clinical trials with plasmid DNA vaccines and are also developing recombinant Ad5 vaccines that will enter clinical trials soon. The relative immunogenicity of these various vaccines will be determined empirically in these trials, and hopefully some of these vaccines will be advanced into phase III efficacy trials. Clinical trials are expensive, challenging, and time-consuming, but they are clearly critical for HIV vaccine development.

Conclusions

While the generation of broadly reactive neutralizing antibodies remains an elusive goal, CTL-based vaccines are actively being evaluated in clinical trials. However, many basic questions still need to be answered in the development of a preventive HIV vaccine. For example, it is important to define which antigens and delivery vectors will elicit the most effective neutralizing antibody and CTL responses. Moreover, strategies will need to be developed to minimize the ability of the virus to escape from neutralizing antibodies and CTL. In addition, immunogen sequences will have to be chosen carefully to cover the diversity of HIV-1 subtypes worldwide most effectively. Finally, the vaccine will need to be manufactured easily and will have to be stable in order to be accessible for people in developing countries. It is likely that an iterative approach will gradually improve the immunogenicity of candidate HIV-1 vaccines over the coming years.

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