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Editorial

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Vaccines against HIV

In the past, empiricism has played a large part in devising new vaccines: effective vaccines have often resulted, in the absence of a precise understanding of how they work. It was accepted by most people that devising vaccines against human immunodeficiency virus (HIV) might be different and would require a detailed knowledge of the biology of the virus, and of the natural course of infection. However, few can have foreseen just how difficult the path to an effective vaccine would prove to be. Here we briefly review the current state of the effort to produce an effective vaccine for HIV.

HIV belongs to the Lentivirus family of retroviruses, which typically establish persistent infections that may lead to slowly progressive disease.[1,2] In addition to the genes for the envelope protein (env), gag precursor and polymerase which all retroviruses carry, lentiretrovirus genomes also code for about five regulatory proteins whose function is the subject of intense study. These viruses show substantial genetic diversity, especially within the envelope protein, as a result of mutations which arise during reverse transcription of the viral genome. Thus an HIV-infected individual acquires a heterogeneous mixture of related but subtly different virus variants which evolves through time;[3] treatment with antiviral drugs can select for the emergence of drug-resistant variants.[4]

Unfortunately, there is no rodent model for HIV-1 vaccine development because primate lentiviruses have a limited host range. HIV-1 can infect only chimpanzees, gibbon apes and pig-tailed macaques. In these primates, it establishes persistent infection but does not induce AIDS. Strains of the related Simian Immunodeficiency Virus (SIV) that naturally infect African sooty mangabeys without causing disease are also capable of infecting Asian rhesus macaques (which are not a natural host), causing in macaques an immunodeficiency disease which closely resembles human AIDS.[5] HIV-2, which is prevalent in West Africa, is genetically closer to SIV than is HIV-1, and is associated with a slower rate of progression to AIDS.

The time from primary infection with HIV-1 until the onset of AIDS is of the order of 10 years or more. During this long asymptomatic phase, infected subjects mount an immune response against the virus. Antibody against different viral components is detectable but is not fully neutralizing, as infectious virus is detectable in plasma at all stages of disease, although the levels of virus are lower during the asymptomatic phase. Cytotoxic T lymphocytes (CTL) specific for HIV are also present at high frequency in the peripheral blood, and appear before antibody during the acute primary infection; their frequency declines later in the course of infection.[6] Memory CTL have also been reported in the peripheral blood of *seronegative* subjects who are at high risk of HIV infection.[7] For all these reasons, CTL are increasingly thought likely to be an important component of the immune response against the virus.

Because lentiviruses integrate their genomes permanently within the host chromosome, the principal goal has been to develop a vaccine which elicits 'sterilizing immunity' capable of eradicating the virus and preventing the establishment of persistent infection. To assess such protective immunity following vaccination, animals are challenged with infectious virus, typically by intravenous injection of 5–10 animal infectious doses of cell-free virus of the same strain used in the vaccine. Because studies of vaccine efficacy can only be carried out in primates, this research is very expensive and the number of animals used in each experiment is usually small ($n=1$–4). Although different vaccines have succeeded in protecting animals against live virus challenge as discussed below, as yet there is only limited information in crucial areas such as the duration of protection conferred by current vaccines, their ability to protect against virus transmitted through mucosal surfaces, and most important, to confer protection against different virus strains. Interestingly, there is evidence that following natural transmission (transplacental or sexual) in human primary HIV-1 infection there is strong selection in favour of certain HIV-1 variants from among a diverse population of virus.[8,9] Immune responses directed against these viruses transmitted in acute primary infection may be crucial to achieve protection in humans.

A variety of different vaccine strategies have been investigated, including vaccines incorporating whole killed virus, live attenuated virus, and genetically engineered subunits of the virus, either as soluble protein or expressed in a replicating vector such as vaccinia virus.[10] In chimpanzees, neither whole killed HIV-1, nor HIV-1 envelope protein expressed in vaccinia followed by booster injections of purified envelope protein, have protected against subsequent challenge with live HIV-1. However, repeated injection of purified HIV-1 envelope protein gp120 succeeded in achieving protection against live virus challenge, apparently through induction of neutralizing antibody which prevents virus from infecting cells.[11] Similarly, passive immunization in chimpanzees by infusion of a strain-specific neutralizing monoclonal antibody also conferred protection against challenge with the same strain of virus.[12] In chimpanzees, it has also been possible to achieve protection against virus-infected cells by vaccination with a complicated schedule of different envelope protein and peptide preparations.[13] Live attenuated HIV-1 vaccines (in which non-essential genes have been deleted from the viral genome) are currently under investigation in chimpanzees.

In the SIV-macaque model, it appeared that whole killed SIV vaccines conferred protection against live SIV challenge.[14,15] However, both the vaccine virus and the live challenge virus had been grown in the same human cell line, and subsequently it became clear that the vaccinated animals had mounted an immune response against *human* cellular proteins (particularly MHC molecules) that had been incorporated into the virus particles in the vaccine, and that this anti-human immunity protected the monkeys against challenge with live pathogenic virus which also contained the same human proteins. Unfortunately, when challenged with live SIV which had been grown in monkey cells, all of these same vaccinated animals became infected.[16] In contrast to the chimpanzee experiments, in macaques repeated injection of SIV envelope protein alone has failed to confer protection. Initial vaccination with live replicating vaccinia expressing SIV envelope protein followed by booster doses of envelope protein has succeeded in protecting macaques from live virus challenge,[17] although this approach has not been universally successful.

The most convincing protection against SIV in macaques has been achieved using a live attenuated virus, from which a non-essential regulatory gene (called nef) had been deleted. *In vitro*, this genetically modified virus shows similar replication to wild-type SIV, but *in vivo* in macaques it establishes a persistent infection with very low levels of viral replication *without* inducing disease. Furthermore, a single dose of the attenuated vaccine virus elicits immunity lasting for at least 2 years which is capable of protecting animals against challenge with up to 1000 animal infectious doses of live pathogenic SIV.[18] The mechanism(s) of this protective immunity are currently under investigation. Notwithstanding the impressive success of this live attenuated vaccine in macaques, and the urgent need to contain the global HIV-1 pandemic, there remain important concerns about the safety of a live attenuated virus capable of integrating into the human genome, and which might possibly revert to the virulent form. The suggestion of performing limited safety testing of a live attenuated HIV-1 in high-risk uninfected human volunteers is highly controversial.

To date, studies in humans have concentrated on the immunogenicity of genetically engineered subunits of HIV-1 and, in particular, the envelope protein gp120, with the intention of inducing neutralizing antibody. Vaccines comprising recombinant envelope protein, with or without initial inoculation of vaccinia virus expressing the envelope protein, have elicited antibodies against the envelope protein, but neutralizing antibody has been directed principally against the vaccine strain, *not* against other laboratory-adapted HIV-1 strains; the titre of neutralizing antibody has been disappointingly low against recent clinical isolates of HIV.[19] As regards induction of cell-mediated immunity, these subunit vaccines can elicit proliferation by $CD4^+$ T helper lymphocytes in response to HIV-1 antigens, and these CD4 cells may be cytotoxic for HIV-infected cells; however, they show little or no induction of $CD8^+$ cytotoxic T lymphocytes which are a feature of the host response in natural infection.[20]

In the light of previous research in animal models, and the failure of sera from vaccine recipients to neutralize clinical isolates, there have been serious doubts about the likely protective efficacy of current recombinant gp120 vaccines, and proposed phase III clinical trials of their efficacy in uninfected volunteers in the USA have recently been suspended.[21,22] As a vaccine for human use seems at least 5 years away, there have been calls for broader support for basic research into HIV-1 pathogenesis,[23,24] including the capacity of some primates to control lentivirus replication successfully without developing disease.

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References

1. McCune JM. HIV-1: the infective process *in vivo*. *Cell* 1991; **64**:351–63.

2. Haase AT. Pathogenesis of lentivirus infections. *Nature* 1986; **322**:130–6.

3. Meyerhans A, Cheynier R, Albert J, Seth M, *et al.* Temporal fluctuations in HIV quasispecies *in vivo* are not reflected by sequential HIV isolations. *Cell* 1989; **58**:901–10.

4. Larder BA, Darby G, Richman DD. HIV with reduced sensitivity to Zidovudine (AZT) isolated during prolonged therapy. *Science* 1989; **243**:1731–4.

5. Desrosiers R. Simian immunodeficiency viruses. *Ann Rev Microbiol* 1988; **42**:607–25.

6. Carmichael AJ, Jin X, Sissons JGP, Borysiewicz LK. Quantitative analysis of the human immunodeficiency type 1 (HIV-1)-specific cytotoxic T lymphocyte (CTL) response at different stages of HIV-1 infection. *J Exp Med* 1993; **177**:249–57.

7. Rowland-Jones SL, Nixon DF, Aldhous MC, Gotch F, *et al.* HIV-specific cytotoxic T-cell activity in an HIV-exposed but uninfected infant. *Lancet* 1993; **341**:860–1.

8. Wolinsky SM, Wike CM, Korber BTM, Hutto C, *et al.* Selective transmission of human immunodeficiency virus type-1 variants from mothers to infants. *Science* 1992; **255**:1134–7.

9. Zhu T, Mo H, Wang N, Nam DS, *et al.* Genotypic and phenotypic characterization of HIV-1 in patients with primary infection. *Science* 1993; **261**:1179–81.

10. Haynes BF. Scientific and social issues of human immunodeficiency virus vaccine development. *Science* 1993; **260**:1279–85.

11. Berman PW, Gregory TJ, Riddle L, Nakamura GR, *et al.* Protection of chimpanzees from infection by HIV-1 after vaccination with recombinant glycoprotein gp120 but not gp160. *Nature* 1990; **345**:622–5.

12. Emini EA, Schleif WA, Nunberg JH, Conley AJ, *et al.* Prevention of HIV-1 infection in chimpanzees by gp120 V3 domain-specific monoclonal antibody. *Nature* 1992; **355**:728–30.

13. Fultz PN, Nara P, Barre-Sinoussi F, Chaput A, *et al.* Vaccine protection of chimpanzees against challenge with HIV-1-infected peripheral blood mononuclear cells. *Science* 1992; **256**:1687–9.

14. Murphey-Corb M, Martin LN, Davison-Fairburn B, Montelaro RC, *et al.* A formalin-inactivated whole SIV vaccine confers protection in macaques. *Science* 1989; **246**:1293–7.

15. Stott EJ, Chan WL, Mills KHG, Page M, *et al.* Preliminary report: protection of cynomolgus macaques against simian immunodeficiency virus by fixed infected-cell vaccine. *Lancet* 1990; **336**:1538–41.

16. Stott EJ. Anti-cell antibody in macaques. *Nature* 1991; **353**:393.

17. Hu S-L, Abrams K, Barber GN, Moran P, *et al.* Protection of macaques against SIV infection by subunit vaccines of SIV envelope glycoprotein gp160. *Science* 1992; **255**:456–9.

18. Daniel MD, Kirchhoff F, Czajak SC, Sehgal PK, Desrosiers RC. Protective effects of a live attenuated SIV vaccine with a deletion in the *nef* gene. *Science* 1992; **258**:1938–41.

19. Cohen J. Jitters jeopardize AIDS vaccine trials. *Science* 1993; **262**:980–1.

20. Walker BD, Chakrabati S, Moss B, Paradis TJ, *et al.* HIV-specific cytotoxic T lymphocytes in seropositive individuals. *Nature* 1987; **328**:345–8.

21. Ada G. Towards phase III trials for candidate vaccines. *Nature* 1993; **364**:489–90.

22. Macilwain C. US puts large-scale AIDS vaccine trials on ice as 'premature'. *Nature* 1994; **369**:593.

23. Fields BN. AIDS: time to turn to basic science. *Nature* 1994; **369**:95–6.

24. Pantaleo G, Graziosi C, Fauci AS. The immunopathogenesis of human immunodeficiency virus infection. *N Engl J Med* 1993; **328**:327–35.