Long-term SHIV Suppression Using AAV Delivery of Monoclonal Antibodies

Jose M. Martinez-Navio[1], Sebastian P. Fuchs[1], Desiree E. Mendes[1], Claudia P. Ramos Muniz[1], Eva G. Rakasz[2], Guangping Gao[3], Jeffrey D. Lifson[4] and Ronald C. Desrosiers[1]

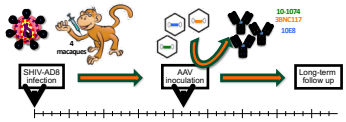
[1] Department of Pathology, Miller School of Medicine, University of Miami, Miami, Florida, USA. [2] Wisconsin National Primate Research Center, University of Wisconsin, Madison, WI, USA. [3] Gene Therapy Center, University of Massachusetts Medical School, Worcester, MA, USA. [4] AIDS and Cancer Virus Program, Frederick National Laboratory for Cancer Research, Frederick, Maryland, USA.

Background

HIV infection can be suppressed with anti-retroviral treatment (ART) but the existence of long-term reservoirs allows the viral load to rebound quickly after ART interruption[1]. Attempts to purge these reservoirs in vivo have not been successful and viral inducers may have serious undesirable effects[2,3]. Consequently, there is intense interest at the current time in finding strategies to prevent this rebound, i.e. finding a "functional cure". Passive transfer of broadly neutralizing antibodies (bNAbs) can prevent infection[4-6], and also suppress active infection in humanized mice and macaques[7-10]. Importantly, bNAbs can also suppress HIV emerging from the viral reservoir[11] and therefore they could be a good alternative to ART[12,13]. However, periodic administrations of large amounts of protein would be required for long-term effects, otherwise and similarly to ART, discontinuation of bNAb therapy would result in viral rebound[12]. A potential solution for overcoming this, is the use of recombinant adeno-associated virus vectors (AAV)[13]. AAV has an outstanding safety record in clinical trials[14] and, as long as the delivered protein is viewed as self[15], it can result in continuous durable expression of the transgene product for years[16-20].
The idea is that HIV-infected people could get one shot of AAV making a cocktail of bNAbs and if satisfactory levels of antibody could be maintained *in vivo*, those individuals would remain suppressed for years without having to take ART or receive regular antibody administrations. Our intention is to perform experiments in monkeys that will inform and guide development of this concept for use in people.

Methods

1st experiment: after 86 weeks of SHIV-AD8 infection, 4 rhesus macaques were inoculated intramuscularly with 3 different AAV1s encoding for 3 different broadly neutralizing anti-HIV antibodies in a therapeutic approach (10-1074, 3BNC117 and 10E8):



2nd experiment: in a similar trial, after 36 weeks of SHIV-AD8 infection, 6 rhesus macaques were inoculated intramuscularly with 4 different AAV8s encoding for 4 different broadly neutralizing anti-HIV antibodies in a therapeutic approach (N6, PGT128, 35O22 and PGT145). They received a booster inoculation with AAV1s encoding for the same antibodies.

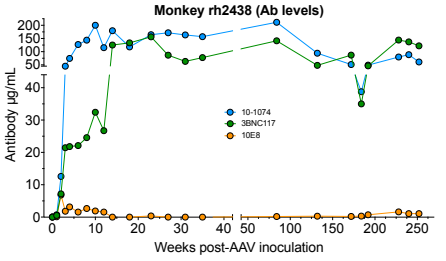
In both experiments viral loads were measured by RT-PCR. Antibody and anti-antibody levels were measured by standard ELISA.

Abstract

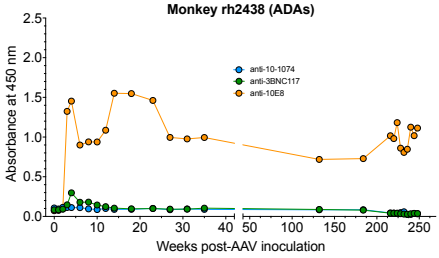
Background: Long-term delivery of anti-HIV monoclonal antibodies using adeno-associated virus (AAV) holds promise for the treatment of HIV infection. We have previously reported monkey rh2438 in which a single administration of AAVs encoding a combination of potent and broadly neutralizing antibodies during the chronic phase of infection resulted in an abrupt decline in plasma viremia which remained below the limit of detection for 38 successive measurements over a 3-year period (Martinez-Navio & Fuchs et al. Immunity 2019). The field has nicknamed this monkey "the Miami monkey" analogous to "the Berlin patient", a person that was cured of his HIV infection. This monkey never received antiviral drugs at any time and therefore appeared functionally cured.
Methods: Indian-origin rhesus macaques (Macaca mulatta) housed at the Wisconsin National Primate Research Center were used for our studies. Monkeys received recombinant AAV vectors expressing full length authentic IgG1 versions of the monoclonal antibodies. Rhesus monkeys were infected with SHIV-AD8 months before receiving AAV expressing constant-region rhesusized versions of select anti-HIV monoclonal antibodies. Antibody and anti-antibody levels were measured by ELISA.
Results: Here we report that monkey rh2438 continues to be suppressed (5 years and counting) and expressing high levels of antibodies 10-1074 and 3BNC117 in serum. Monkey rh2438 generated little or no anti-drug antibodies to these antibodies. Additionally, we have two other macaques, r14121 and r14097, which also received AAVs coding for a cocktail of neutralizing anti-HIV antibodies during the chronic phase of infection and have shown suppressed viral loads for the last 2 years. Monkey r14121 showed sustained delivery of reasonable levels of antibody PGT128 and a late rise of antibody N6. Monkey r14097 showed sustained delivery of antibodies PGT128 and N6 at reasonable levels and a late rise of antibodies 35O22 and PGT145.
Conclusions: Our data show that durable, continuous antibody expression can be achieved after one administration of AAV and support the potential for lifelong suppression of viral loads with the AAV-antibody approach.

Results

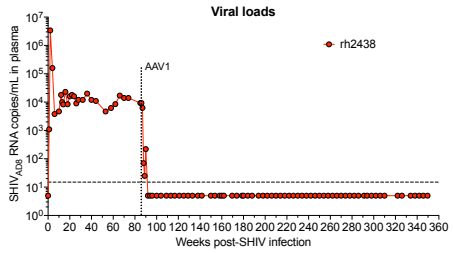
1. Monkey rh2438 persistently maintained high concentrations of antibodies 3BNC117 and 10-1074 in serum following the AAV administration with a late rise of antibody 10E8:



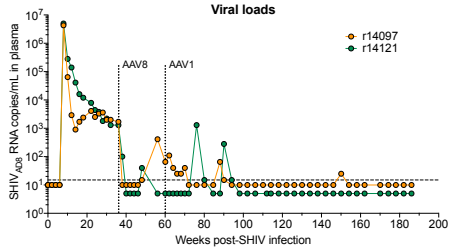
2. Monkey rh2438 generated little or no anti-drug antibodies (ADAs) to the 10-1074 and 3BNC117 antibodies.



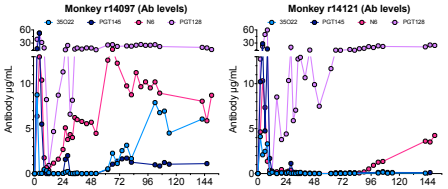
3. After AAV administration, animal rh2438 achieved profound and sustained virologic control during the chronic phase of the infection:



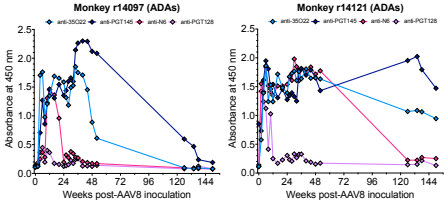
4. In a second therapy trial, monkeys r14097 and r14121 exhibited suppression of viral loads after AAV therapy with antibodies:



5. Monkey r14097 showed sustained delivery of two antibodies at reasonable levels: PGT128 (20-25 µg/ml) and N6 (5-15 µg/ml); and a late rise of antibody 35O22 to 5-8 µg/ml and of antibody PGT145 to 1 µg/ml. Monkey r14121 showed sustained delivery of 10-20 µg/ml of antibody PGT128, and a late rise of antibody N6 to 4 µg/ml:



6. ADAs in monkeys r14097 and r14121 seemed to explain well the antibody levels:



References

1. Chun, T.W. and A.S. Fauci, HIV reservoirs: pathogenesis and obstacles to viral eradication and cure. AIDS, 2012. 26(10): p. 1261-8.
2. Archin, N.M., et al., Eradicating HIV-1 infection: seeking to clear a persistent pathogen. Nat Rev Microbiol, 2014. 12(11): p. 750-64.
3. Shan, L., et al., Stimulation of HIV-1-specific cytolytic T lymphocytes facilitates elimination of latent viral reservoir after virus reactivation. Immunity, 2012. 36(3): p. 491-501.
4. Shingai, M., et al., Passive transfer of modest titers of potent and broadly neutralizing anti-HIV monoclonal antibodies block SHIV infection in macaques. J Exp Med, 2014. 211(10): p. 2061-74.
5. Saunders, K.O., et al., Sustained Delivery of a Broadly Neutralizing Antibody in Nonhuman Primates Confers Long-Term Protection against Simian/Human Immunodeficiency Virus Infection. J Virol, 2015. 89(11): p. 5895-903.
6. Moldt, B., et al., Highly potent HIV-specific antibody neutralization in vitro translates into effective protection against mucosal SHIV challenge in vivo. Proc Natl Acad Sci U S A, 2012. 109(46): p. 18921-5.
7. Klein, F., et al., HIV therapy by a combination of broadly neutralizing antibodies in humanized mice. Nature, 2012. 492(7427): p. 118-22.
8. Barouch, D.H., et al., Therapeutic efficacy of potent neutralizing HIV-1-specific monoclonal antibodies in SHIV-infected rhesus monkeys. Nature, 2013. 503(7475): p. 224-8.
9. Shingai, M., et al., Antibody-mediated immunotherapy of macaques chronically infected with SHIV suppresses viraemia. Nature, 2013. 503(7475): p. 277-80.
10. Horwitz, J.A., et al., HIV-1 suppression and durable control by combining single broadly neutralizing antibodies and antiretroviral drugs in humanized mice. Proc Natl Acad Sci U S A, 2013. 110(41): p. 16538-43.
11. Chun, T.W., et al., Broadly neutralizing antibodies suppress HIV in the persistent viral reservoir. Proc Natl Acad Sci U S A, 2014. 111(36): p. 13151-6.
12. Halper-Stromberg, A., et al., Broadly neutralizing antibodies and viral inducers decrease rebound from HIV-1 latent reservoirs in humanized mice. Cell, 2014. 158(5): p. 989-99.
13. Deal, C.E. and A.B. Balazs, Vectored antibody gene delivery for the prevention or treatment of HIV infection. Curr Opin HIV AIDS, 2015. 10(3): p. 190-7.
14. Hastie, E. and R.J. Samulski, Adeno-associated virus at 50: a golden anniversary of discovery, research, and gene therapy success-a personal perspective. Hum Gene Ther, 2015. 26(5): p. 257-65.
15. Martinez-Navio, J.M., et al., Host Anti-antibody Responses Following AAV-Mediated Delivery of Antibodies against HIV and SIV in Rhesus Monkeys. Mol Ther, 2015.
16. Daya, S. and K.I. Berns, Gene therapy using adeno-associated virus vectors. Clin Microbiol Rev, 2008. 21(4): p. 583-93.
17. Goncalves, M.A., Adeno-associated virus: from defective virus to effective vector. Virol J, 2005. 2: p. 43.
18. McCarty, D.M., Self-complementary AAV vectors; advances and applications. Mol Ther, 2008. 16(10): p. 1648-56.
19. Schultz, B.R. and J.S. Chamberlain, Recombinant adeno-associated virus transduction and integration. Mol Ther, 2008. 16(7): p. 1189-99.
20. Mingozzi, F. and K.A. High, Therapeutic in vivo gene transfer for genetic disease using AAV: progress and challenges. Nat Rev Genet, 2011. 12(5): p. 341-55.

Conclusions

1. **Durable, continuous antibody expression can be achieved after one administration of AAV.**
2. **Our data support the potential for lifelong suppression of viral loads with the AAV-antibody approach.**