Nussenzweig: Development of optimized broadly neutralizing antibodies for HIV-1 prevention

OVERVIEW

Despite development of excellent therapeutics and strategies for HIV-1 prevention, HIV/AIDS infection remains a major problem in both the developed and developing world. Vaccines do not exist and there are significant barriers to their development. Although not considered practical until very recently, the discovery of highly potent broadly neutralizing antibodies has brought new impetus to the passive antibody approach to HIV-1 prevention. Importantly these antibodies have been shown to prevent infection in mice and macaque models and have shown significant efficacy against HIV-1 in a phase 1 clinical trial. A key objective of this proposal is to improve the activity of the naturally occurring anti-HIV-1 antibodies by altering the mechanism of antigen recognition and/or by enhancing their effector functions. The majority of the research activities run in parallel, as the main aim of the proposal is to continuously generate optimized anti-HIV-1 antibodies and evaluate their activity in in vivo infection models using humanized mice and NHP. These activities are expected to occur throughout the entire 4-year period and it is anticipated that every year new anti-HIV-1 antibodies will be isolated, optimized and eventually tested in vivo. By the end of the 4-year period, new classes of anti-HIV-1 antibodies with substantially improved neutralization activity and effector function will be available for pre-clinical evaluation in humans. The 4-year period for the grant reflects the requirement that the majority of the described in vivo experiments, and particularly those involving NHP, require a minimum of least a year to complete.

This collaborative effort is led by Michel Nussenzweig, MD, PhD (The Rockefeller University) with Jeffery Ravetch, MD, PhD (The Rockefeller University) as Co-Principal Investigator, and the participation of Pamela Bjorkman, PhD (California Institute of Technology) and Malcolm Martin, MD (National Institute of Allergy and Infectious Diseases). The award was received in June, 2015 with an initial agreement length of 4 years.

Grant at a Glance

Principal Investigator
Michel Nussenzweig, MD, PhD

Grantee Institution
The Rockefeller University, New York, USA

Project Title
Development of optimized broadly neutralizing antibodies for HIV-1 prevention

OPPID
1124068

Grant Award
Up to $13 million, awarded in June, 2015

Collaborating Institutions
- ◊ California Institute of Technology, Pasadena, USA
- ◊ National Institute of Allergy and Infectious Diseases – Viral Pathogenesis and Vaccine Section, Bethesda, USA

RESEARCH OBJECTIVES

1.) Optimized antibody based therapeutic interventions for HIV-1 prevention in pre-clinical models for translation to humans
   - 1.1.) Produce humanized mice for the studies proposed by all investigators in the consortium
   - 1.2.) Optimize antibody-based interventions for prevention in preclinical models

2.) Develop optimized anti-HIV-1 bnAbs with increased potency/breadth and resistance to viral mutation
   - 2.1.) Improve conventional format human IgG antibodies using structure-based design
   - 2.2.) Create intra-spike crosslinking reagents that utilize avidity effects to neutralize with synergy
   - 2.3.) Produce most potent/most synergistic bispecific reagents for *in vivo* evaluation

3.) Generation and pre-clinical evaluation of Fc-optimized anti-HIV-1 antibodies with improved effector activity
   - 3.1.) Assess *in vivo* activity of Fc optimized anti-HIV-1 bnAbs in humanized mouse models of HIV-1 infection
   - 3.2.) Evaluate *in vivo* protective activity of Fc optimized bnAbs in non-human primates

4.) Blocking the establishment of virus infection with anti-HIV-1 bnAbs using the SHIV macaque model of HIV AIDS
   - 4.1.) Assess *in vivo* potency and durability of optimized anti-HIV Nabs in pre-exposure prophylaxis experiments
   - 4.2.) Suppress virus replication in SHIV infected macaques

PROGRESS

1.) This sub-objective focuses on producing mice for testing bNAb-based prevention strategies. It requires breeding NOD/SCID γc-/- mice, and identifying newborn mice to use as recipients, production of CD34+ fetal liver cells for transfer, irradiation and injection of recipients and tracking the level of reconstitution by flow cytometry. The mice have been used to assay antibody activity including prevention and therapy in our own laboratory and in collaborative experiments with Dr. Bjorkman and Dr. Ravetch. To date the mouse experiments have been predictive of antibody activity in humans. In addition we have developed new assays for antibody clearance of infected cells in humanized mice. These experiments, recently published in Science, revealed that Fc effector functions contribute to antibody activity in humans by accelerating the clearance of infected cells.

2.) We solved a 3.5Å crystal structure of a native-like Env trimer with fully-processed native glycosylation, revealing an extensive glycan shield of untrimmed high-mannose and complex-type N-glycans on an Env crystal structure for the first time. The trimer was complexed with two glycan-interacting bNAbs from the Nussenzweig lab: 10-1074 against the V3-loop (in clinical trials), and IOMA, a new CD4bs antibody. Although IOMA evolved from the VH1-2*02 germline gene of CD4bs-targeting VRC01-class bNAbs, its light chain lacks the short (5 aa) CDRL3 loop that defines VRC01-class bNAbs, and its binding resembles orientations of VH1-46–derived CD4bs bNAbs. The existence of bNAbs that mix features of VRC01-class and VH1-46-class antibodies has implications for immunization strategies targeting VRC01-like bNAbs; namely, researchers should search for VH1-2–derived Abs with normal-length CDRL3s in vaccine results (instead of focusing only on VH1-2 derived Abs with 5-residue CDRL3s) because these IOMA-like bNAbs would be easier to elicit than VRC01-class bNAbs since they are less heavily mutated.

3.) A panel of Fc domain variants of human IgG1 has been generated with differential binding capacity to the different classes of human and mouse FcγRs. Out of these variants, GASDALIE (G236A/S239D/A330L/I332E) exhibited the best enhancement in the affinity for activating FcγRs, without any impairment for FcRn binding. This variant was selected and bNAb GASDALIE Fc variants were generated for 3BNC117,

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10-1074 and PG16 and their activity assessed in in vivo experiments using HIV-1-infected humanized mice. In preliminary experiments, we have assessed the activity of Fc-optimized bNAbs (3BNC117, 10-1074 and PG16) in HIV-1-infected humanized mice. Compared to wild-type hIgG1 or FcRnull binding bNAbs, Fc-optimized (GASDALIE) bNAbs revealed improved in vivo therapeutic activity.

We have assessed the capacity of wild-type and Fc-optimized anti-HIV-1 bNAbs to engage rhesus macaque FcγRs. Macaque FcγR were cloned and their extracellular, IgG binding domain was expressed recombinantly. Their binding affinity for wild-type hIgG1 and Fc domain variants was measured by surface plasmon resonance (SPR). Additionally, genetic variants of rhesus macaque FcγRs were analyzed and their impact on human IgG1 binding was characterized. No significant impact on their binding capacity for hIgG was observed for the most common genetic variants of rhesus FcRs.

4.) Based on the relatively long term protection conferred by Hepatitis A immune globulin, we tested the efficacy of a single injection (20mg/kg) of four anti-HIV-1 neutralizing monoclonal antibodies (MAbs) (VRC01, VRC01-LS, 3BNC117, and 10-10749-12) in blocking repeated weekly low dose virus challenges of the clade B SHIVAD8. Compared to control animals, which required 2 to 6 challenges (median=3 weeks) for infection, a single bNAb infusion prevented virus acquisition for up to 23 weeks. This effect depended on antibody potency and half-life. The highest levels of plasma neutralizing activity and correspondingly, the longest protection, were found in monkeys administered the more potent antibodies, 3BNC117 and 10 1074 (median=13 and 12.5 weeks respectively). VRC01, which showed lower plasma-neutralizing activity, protected for a shorter time (median=8 weeks). The introduction of a mutation that extends antibody half-life into the Fc domain of VRC01 increased median protection from 8 to 14.5 weeks.