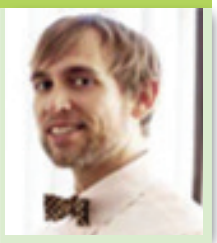
Ward: Establishment of an Analytics Network to Support Qualified/Validated Assays Required for Characterization of HIV Env Immunogens

OVERVIEW

Eliciting broadly neutralizing antibodies (bnAbs) that target the envelope glycoprotein (Env) spike on the surface of HIV-1 is thought to be required for a successful HIV vaccine. Env is a highly glycosylated trimer of heterodimers composed of gp120 and gp41 subunits. While a large number of bnAbs from naturally infected patients have been isolated and characterized, little is known regarding how to elicit such antibodies with a vaccine. A number of recent efforts have been placed on developing trimeric Env proteins that preferentially expose bnAb epitopes while masking non-nAb epitopes, similar to the native spike. This task has proven quite difficult because Env is a meta-stable type I viral fusion machine that has a large number of disulfide bonds that must be correctly formed during the folding process.

We have demonstrated that a well-folded, native-like trimer has implications for antigenicity, immunogenicity, glycosylation pattern, and thermostability. It requires several assays to accurately characterize these qualities of Env trimers. These include i) differential scanning calorimetry (DSC) to assess stability, (ii) bio-layer interferometry (Octet) to assess antigenic profile, (iii) antibody capture and size exclusion chromatography to assess purity, (iv) negative stain EM to assess conformational and compositional heterogeneity and antigenicity, and (v) high-resolution cryoEM to assess the molecular details of Env trimers. Moreover, we conduct such assays with rigorous controls/comparators in order to avoid misinterpretation and flawed conclusions. Overall, we have developed a pipeline with these methods to rapidly analyze and compare Env trimer candidates, furnishing results in a timely manner as Env Immunogens are devised and produced for immunization programs. These data can then be directly compared to the outcomes from animal immunization experiments, thereby informing rational vaccine design and facilitating iteration between structure-based Env design and empirical vaccination. We have adapted our imaging pipeline and leveraged our considerable database of trimer-antibody images to map polyclonal antibody responses during vaccine trials. We and others are using this information to guide immunogen redesign with the goal of guiding immune responses toward development of broadly neutralizing antibodies.

Grant at a Glance



Principal Investigator
Andrew Ward

Grantee Institution
The Scripps Research Institute, La Jolla, USA

Project Title
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Grant Award
Up to $3.6 million, awarded in June, 2015

We also conduct the above assays using adjuvant formulated immunogens, conditions that we refer to as stress tests, in order to ensure high quality reagents are delivered to animal and human immunization experiments. This includes a recently produced cGMP batch of BG505 SOSIP.664 trimers suitable for human clinical trials. Finally, we have begun to construct and characterize Env trimer presenting nanoparticles to potentially improve immunogenicity.

This grant is led by Andrew Ward, PhD (The Scripps Research Institute). The award was received in June, 2015 with an initial agreement length of 5 years.

RESEARCH OBJECTIVES

1.) To perform structural and biophysical characterization on up to 1000 Env trimers and/or trimer antibody complexes per year.

2.) To produce a variety of full-length Env trimers for structural, antigenic, and glycan profile characterization that will serve as important comparators to the soluble, native-like Env trimers being developed as immunogens.

3.) To provide structural information and support for CAVD vaccine design efforts, e.g. the “Nearest Neighbor” consortium.

RESEARCH PROGRESS

1.) In the first three years of the award we have analyzed almost 2300 Env immunogens and Env-antibody complexes from an international network of over 30 collaborating labs from academic and industrial partners. ~100 of these samples have been from the “Nearest Neighbor” consortium and include Env trimer presenting nanoparticles.

2.) This work has resulted in nearly 60 publications and nearly 200 EMDB structure depositions ranging in resolution from 3.3 Å to ~20 Å.

3.) We have supported production and characterization of Env trimer immunogens for many animal immunization experiments and are involved in developing the GMP process for production of trimer immunogens for human vaccine trials.

4.) We have also developed and implemented a real-time EM based analysis of the polyclonal response in ongoing vaccine trials.