# How Does HIV Env Structure Informs Vaccine Design?

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## Background

The DHVI Division of Structural Biology seeks to use atomic level structural information for design of an effective HIV-1 vaccine. Through visualization of the HIV-1 envelope (Env) and its interactions with the human immune system, we obtain structural information that we translate into the rational development vaccine immunogens

## Methods

We use negative stain electron microscopy (NSEM), cryo-electron microscopy (cryo-EM), and x-ray crystallography as the major structural techniques for visualization of HIV-1 Env, and combine these with biochemical and biophysical studies, as well as computational methods to obtain a basic understanding of the functions and interactions of the HIV-1 Env.

## Results:

1. ***The DHVI NSEM pipeline*** runs on a daily basis to quality control vaccine immunogens for animal studies and other applications. Offering rapid sample turnover and economical operations, the NSEM pipeline is the most widely utilized resource of the DHVI Division of Structural Biology. Over the last year, the NSEM team has focused efforts on improving operational speed and data processing allowing high-quality visualization of a large variety of samples including HIV-1 Env immunogens, antibodies, nanoparticles, and VLPs. In the last year we have also expanded our NSEM studies to the analyses of serum samples and mucosal fluids.

2. To understand the ***mechanism of HIV-1 entry*** we have determined structures of HIV-1 entry intermediates. We have determined a 3.8 Å resolution structure of a single CD4 bound to a closed HIV-1 Env trimer revealing new contacts of CD4 with Env. We have also structurally characterized an Env designed to prevent CD4-induced rearrangements by targeted disruption of an allosteric network modulating Env conformational changes.

3. We have structurally characterized the HIV-1 glycan-V3 targeting ***DH270 Broadly Neutralizing Antibody Lineage***. The structures revealed movements in the V1 loop and interactive glycans, shifts in antibody orientations, antibody VH-VL orientations, and antibody elbow angles, as the lineage progressed to maturation.

4. We have solved a structure in complex with the HIV-1 Env immunogen $Man_5$-enriched CH505.N279K.G458Y.SOSIP.664 of the unmutated common ancestor (UCA) of the HIV-1 CD4-binding site targeting ***CH235 Broadly Neutralizing Antibody Lineage***. The structure revealed interactions of the N279K and G458Y mutations with the CDR L3 loop of CH235 UCA thus providing a structural understanding of the role of these mutations in facilitating binding to the CH235 UCA. (see also Henderson *et al* abstract)

5. Using NSEM and cryo-EM we have characterized the structural properties of a novel class of 2G12-mimetic, yet ***non domain-swapped Fab dimer glycan-reactive (FDG) antibodies***. These studies showed that the Fab-dimerized 2G12-like motif is more common than previously thought,

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and that creation of a Fab-dimerized paratope for an HIV-1 neutralizing antibody does not require VH domain-swapping.

6. Finally, the structural team is an integral part of the CHAVD Kalma Immunogen Design Team, wherein we are defining the structural basis of bnAb affinity maturation to guide sequential immunogen design.

**Conclusions:** These results highlight the power of structural information on HIV-1 vaccine design, from leveraging a basic understanding of HIV-1 entry mechanism for immunogen design, to rapid visualization of Env immunogens by NSEM for quality control, discovery of novel antibody interactions, and atomic level visualization of antibody/Env interactions.

References

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