Sequential SHIV-Env Clones with Neutralization Sensitivity for Breadth Development

Manxue Jia, Cecilia Cheng-Mayer, Xueling Wu

Aaron Diamond AIDS Research Center, Affiliate of the Rockefeller University, New York, New York, USA

Background

Neutralizing antibodies gradually increase in breadth in selected SHIV-infected rhesus macaques (RMs) and HIV-1-infected humans. However, the envelope (Env) clones that are responsible for the development of neutralization breadth have not been identified.

Methods

Because the SHIV-RM is the best available animal model for HIV-1 antibody studies, we screened 13 viremic RMs that were intra-rectally infected with the R5 clade B SHIV$_{SF162P3N}$ for neutralization activity against eight HIV-1 heterologous strains. Two animals, GB40 and FF69, were identified as containing the best neutralization breadths and titers. Using plasmas collected longitudinally from these animals, we tested four homologous Env clones derived from the SHIV$_{SF162P3N}$ inoculum and identified two sequential homologous neutralization activities (waves) prior to a third wave that cross-neutralized the tested HIV-1 strains. Based on the time when each wave appears, we selected time points for Env isolation. Full-length *env* sequences were obtained by single genome amplification, and representative Env clones were tested for neutralization.

Results

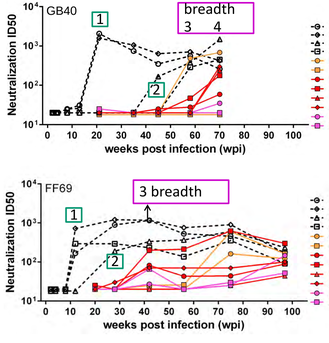


Figure 1. Neutralization breadth development in GB40 and FF69. In both RMs there are at least two waves of homologous responses before appearance of cross-reactivity to heterologous strains. In GB40, the earliest wave appeared at w21, wave 2 at w45, and bnAb activity at w58. In FF69, the earliest wave appeared at w12, wave 2 at w28, and bnAb activity at w42.

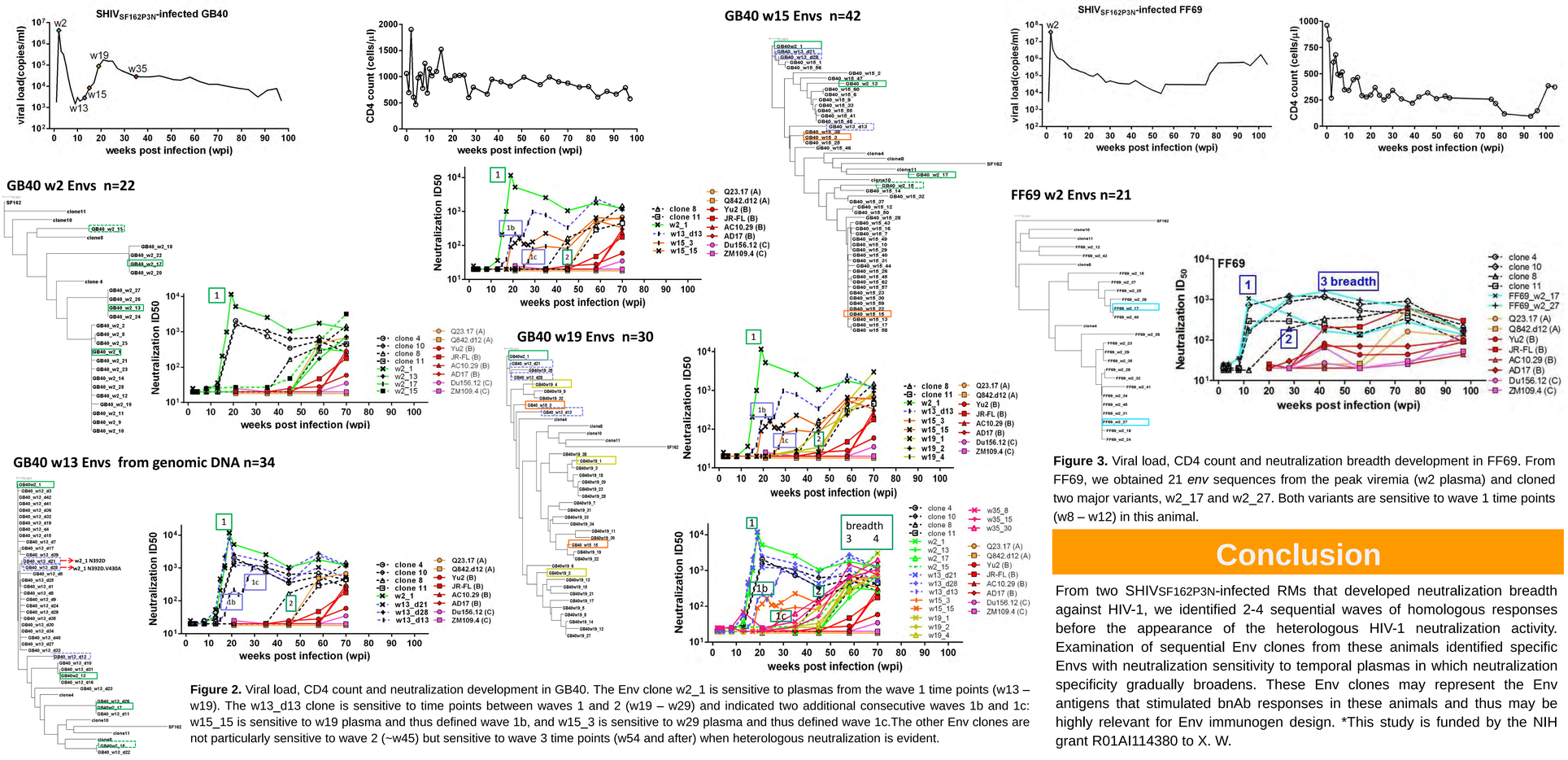


Figure 2. Viral load, CD4 count and neutralization development in GB40. The Env clone w2_1 is sensitive to plasmas from the wave 1 time points (w13 – w19). The w13_d13 clone is sensitive to time points between waves 1 and 2 (w19 – w29) and indicated two additional consecutive waves 1b and 1c: w15_15 is sensitive to w19 plasma and thus defined wave 1b, and w15_3 is sensitive to w29 plasma and thus defined wave 1c.The other Env clones are not particularly sensitive to wave 2 (~w45) but sensitive to wave 3 time points (w54 and after) when heterologous neutralization is evident.

Figure 3. Viral load, CD4 count and neutralization breadth development in FF69. From FF69, we obtained 21 *env* sequences from the peak viremia (w2 plasma) and cloned two major variants, w2_17 and w2_27. Both variants are sensitive to wave 1 time points (w8 – w12) in this animal.

Conclusion

From two SHIV$_{SF162P3N}$-infected RMs that developed neutralization breadth against HIV-1, we identified 2-4 sequential waves of homologous responses before the appearance of the heterologous HIV-1 neutralization activity. Examination of sequential Env clones from these animals identified specific Envs with neutralization sensitivity to temporal plasmas in which neutralization specificity gradually broadens. These Env clones may represent the Env antigens that stimulated bnAb responses in these animals and thus may be highly relevant for Env immunogen design. *This study is funded by the NIH grant R01AI114380 to X. W.