Abstract

A prophylactic HIV-1 vaccine will likely need to elicit broadly neutralizing antibodies (bNAbs) against conserved HIV-1 envelope epitopes such as the gp120-gp41 interface which includes the FP. The isolation of gp120-gp41 interface-, and FP-directed bNAbs from chronically HIV-infected donors has made this epitope an appealing vaccine target. Moreover, promising preclinical immunogenicity animal studies have shown the possibility of eliciting such responses in animals. However, little is known about the population prevalence or kinetics of gp120-gp41 interface responses, including FP-directed responses. Lastly, few FP-directed antibodies have been isolated from people living with HIV (PLWH), limiting our understanding of common developmental pathways that can be explored for vaccine purposes.

Here, we first assessed the prevalence of bNAbs in participants previously enrolled in the CAPRISA 004 Tenofovir gel trial (CAP004). We show that in this cohort, only 12% of individuals developed breadth at three years post-infection, and that high viral load and low CD4 count were associated with bNAb development, as previously reported. ELISA screening showed that only 13% of individuals developed FP-directed responses at three years post-infection. Of the 13% (n=9), only two donors had broad plasma responses, including donor CAP312, whose plasma exhibited 64% neutralization breadth at three years post-infection. In CAP312, FP binding and heterologous neutralization against a multi-clade 22 virus panel appeared simultaneously, within one year (~ 50 weeks post-infection). Taken together, our findings suggest that gp120-gp41 interface- and FP-directed responses are infrequently elicited during infection.

As few FP-specific bNAbs have been isolated to date, little is known about their shared features that could be exploited for eliciting FP-specific bNAbs by vaccination. We next isolated three FP-specific mAbs from donor CAP312 at three years post-infection; AIRU-F8,

AIRU-G9 and AIRU-G4 with 64, 45 and 5% breadth, respectively. We showed that our mAbs, and previously isolated bNAbs PGT151 and ACS202 share gene usage and have a similar unusually long CDRH3, as a result of a conserved motif inherited from the germline *IGHJ6*02* gene. Furthermore, we showed that CAP312 mAbs have even longer CDRH3s compared to PGT151 and ACS202 despite this shared motif. We also showed, using point mutants and glycan mutants that our FP-specific mAbs have a unique neutralization profile compared to published mAbs. Overall, these results suggest that FP-specific mAbs share structural and genetic features that could be explored further for lineage vaccine development.

Lastly, we delineated the ontogeny of the gp120-gp41 interface-directed nAb CAP248-2B by tracing the evolutionary pathways utilising longitudinal samples from 11-281 weeks post-infection (wpi). CAP248-2B interacts with the HIV-1 Env trimer through a long light chain CDRL3 that inserts into the viral membrane, and a heavy chain CDRH1 ^{32}ED^{33} motif that interacts with gp41. We showed that the unusually long light chain CDRL3 and the heavy chain ^{32}ED^{33} motif are functionally redundant against heterologous viruses. We also showed that affinity maturation mutations in this lineage selected a lineage with limited heterologous neutralization breadth.

In summary, these findings support further research into gp120-gp41- and FP-specific responses in multiple cohorts. Furthermore, future studies should aim to elucidate mechanisms governing the development of these responses so that productive developmental pathways can be identified and exploited for vaccine design.