Phenotypic neutralization sieve analysis of an SIV nonhuman primate vaccine challenge study.

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Abstract #349

Abstract

Background: We previously characterized the neutralization sensitivity of the nonhuman primate (NHP) challenge stock SIVsmE660 and found the isolate was generally highly neutralization sensitive with a minor population of resistant variants (Lopker et al., J Virol 2013). We hypothesized that incomplete vaccine-mediated protection may result from protection against only the neutralization sensitive fraction of SIVsmE660. Subsequently, a large NHP vaccine study (VRC-10-332) demonstrated incomplete protection from SIVsmE660 challenge and identified genotypic signatures associated with neutralization resistance (Roederer et al., Nature 2014). Here, we perform ***a phenotypic neutralization sieve analysis*** of the breakthrough Transmitted/Founder TF viruses of this NHP vaccine study.

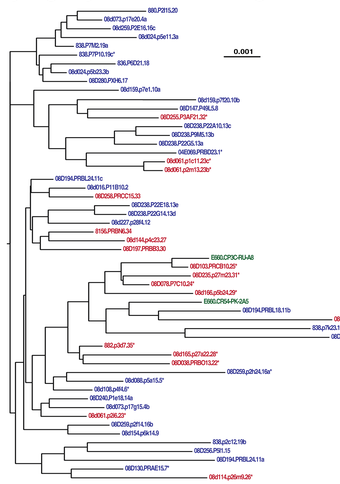
Methods: TF Env clones were derived by single genome sequencing of peak plasma vRNA from the breakthrough infections in the heterologous Env-immunized animals (n=18 TF Envs) and controls (n=35 TF Envs). Neutralization sensitivity was assessed in a TZM-bl assay with pre-challenge immune plasmas from SIVmac239-vaccinated study animals, SIV-infected macaque plasmas and a panel of 10 monoclonal antibodies (mAb) targeting V1, V2, V3, V4, CD4bs and CD4i epitopes.

Results: The breakthrough TF Envs from immunized animals were significantly more neutralization resistant to vaccinated macaque plasma, SIV-infected macaque plasmas and mAbs targeting diverse gp120 epitopes than those from control animals, as measured by IC_{50}, maximal neutralization, and fraction of highly-resistant viruses. The median maximum neutralization of control vs. Env-vaccinated were 69% vs. 24%, 68% vs. 29%, and 73% vs. 26%, respectively for immune plasma, SIV-infected plasma, and anti-CD4bs mAb (all *p<0.0001, Mann-Whitney). The majority of vaccine TF Envs (13/18, 72%) were highly neutralization resistant to polyclonal plasmas and mAbs; only rare Envs were resistant in the control TF Envs (6/35, 17%, p=0.0002) and the SIVsmE660 isolate (10% p=0.0002).* The genotypic signature of resistance identified in t was highly predictive of the phenotypic neutralization sensitivity

Conclusions: We demonstrate a phenotypic neutralization siev NHP vaccine challenge study with substantial enrichment for ne resistant viruses in the breakthrough infections of vaccinated ar findings support our hypothesis that incomplete vaccine-mediat results from selection against the highly neutralization sensitive SIVsmE660 isolate. Further, we corroborate the genetic signat neutralization sensitivity predicted in the parent study. Togethe demonstrate proof-of-concept that phenotypic neutralization sie useful in assessment of vaccine trial outcomes and correlates a

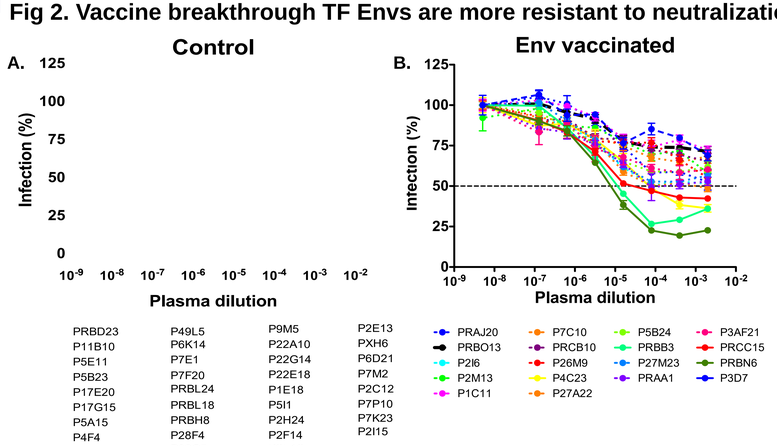
Results

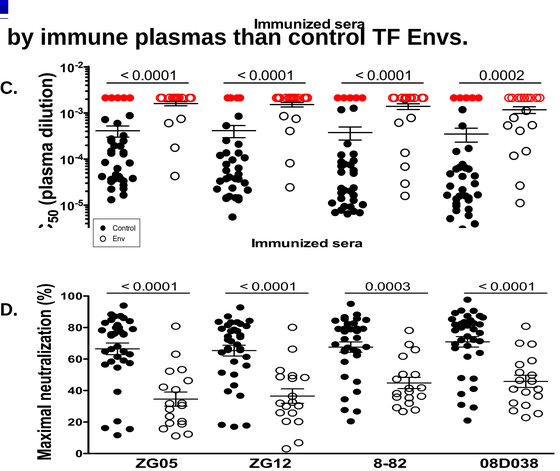
Fig 1. Phylogeny of breakthrough TF Envs



Maximum likelihood phylogeny of the breakthrough TF Envs from the SIVmac239 Env-vaccinated (red) and control (blue) animals, with reference SIVsmE660 TF Envs (green). C1 genetic signature A/K indicated with asterisk.

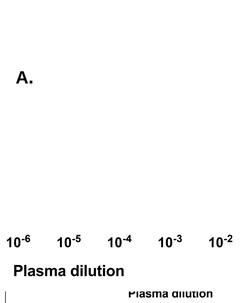
Fig 2. Vaccine breakthrough TF Envs are more resistant to neutralization by immune plasmas than control TF Envs.

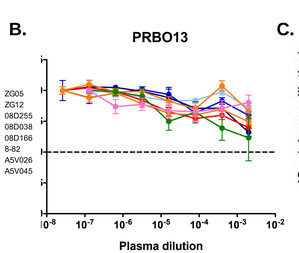


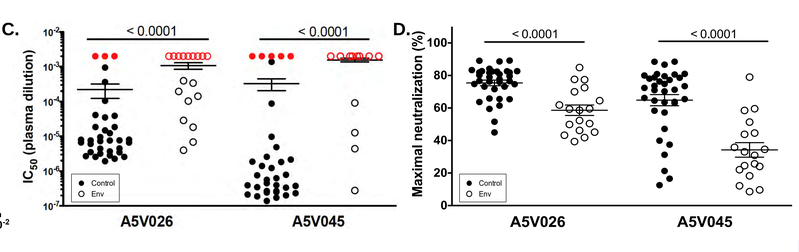


The 53 breakthrough TF Envs (n=35 from control animals (A), n=18 from Env-vaccinated animals (B) were tested for neutralization sensitivity to Env-vaccinated immune plasmas from 4 study animals (ZG05, ZG12, 8-82, and 08D038). The immune plasmas came from two animals that were protected throughout the course of the 12-challenge study (ZG05 and ZG12) and two animals that became infected (8-82 at challenge 7, 08D038 at challenge 2). The TF Envs from the Env-vaccinated animals were significantly more resistant than the TF Envs from control animals, measured by IC_{50} (C) and [illegible] representative neutralization curves from plasma 08D038, where solid lines indicate C1 signature TR, dashed lines indicate C1 signature A/K. In Fig 1C,

TF Envs are more resistant to neutralization by SIV-infected macaque plasmas than controls.

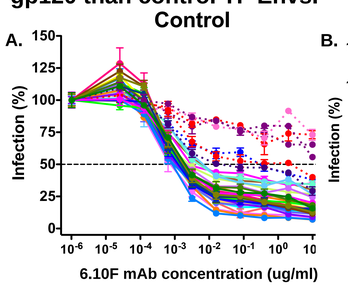


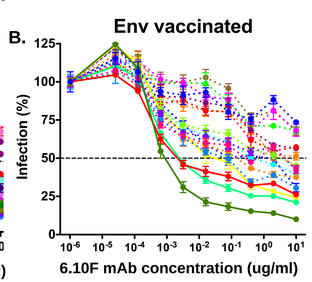




The Env-vaccinated breakthrough TF Envs were more resistant than control TF Envs [illegible] SIV Env mAbs targeting epitopes throughout gp120. Neutralization curves of representative anti-V3 mAb 6.10F to Control (4A) and SIVmac239 Env-vaccinated TF Envs (4B). Maximal neutralization at high concentration of mAb was significantly greater in control vs. Env-vaccinated Envs for all 10 mAbs (4C).

Fig 4. Vaccine breakthrough TF Envs are *globally* more resistar gp120 than control TF Envs.





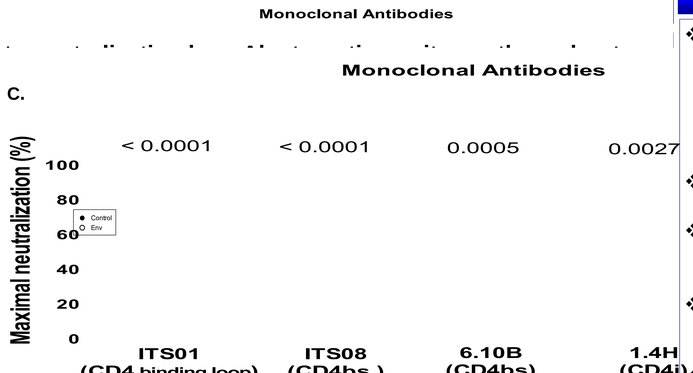
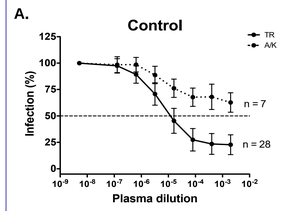
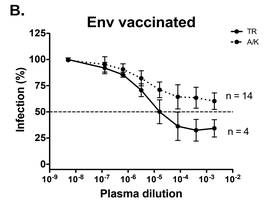
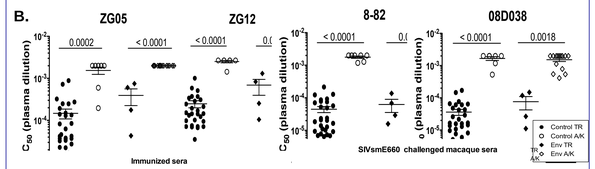
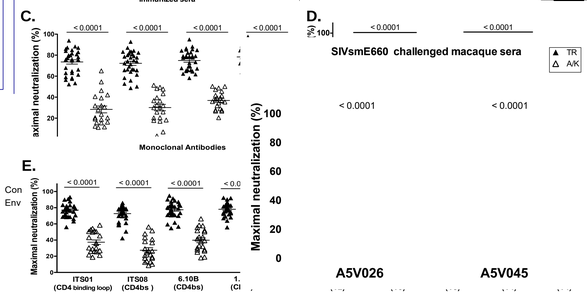


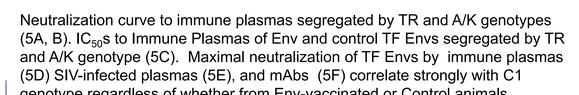
Fig 5. The 2 AA genetic signature in C1 identified by Roederer, et al. was highly predictive of phenotypic neutralization resistance.











Neutralization curve to immune plasmas segregated by TR and A/K genotypes (5A, B). IC_{50}s to Immune Plasmas of Env and control TF Envs segregated by TR and A/K genotype (5C). Maximal neutralization of TF Envs by immune plasmas (5D) SIV-infected plasmas (5E), and mAbs (5F) correlate strongly with C1 genotype regardless of whether from Env-vaccinated or Control animals.

Conclusions

- We demonstrate a phenotypic neutralization sieve effect in a large NHP vaccine challenge study. The SIVmac239 Env-vaccinated breakthrough TF Envs were significantly more neutralization resistant than controls to immune plasma, SIV-infected macaque plasmas, and mAbs targeting gp120 epitopes.
- The C1 genetic signature (TR vs. A/K) was highly predictive of phenotypic neutralization sensitivity.
- Findings demonstrate proof-of-concept that phenotypic neutralization sieve analyses can aid assessment of vaccine trial outcomes and correlates analyses.
- Findings support the utility of heterogeneous challenge swarms to determine the threshold of vaccine induced protection.
- The unique neutralization properties of SIVsmE660 (extreme, global sensitivity of majority of swarm, plateau of retained infectivity in resistant viruses) hinder its ability to recapitulate HIV.