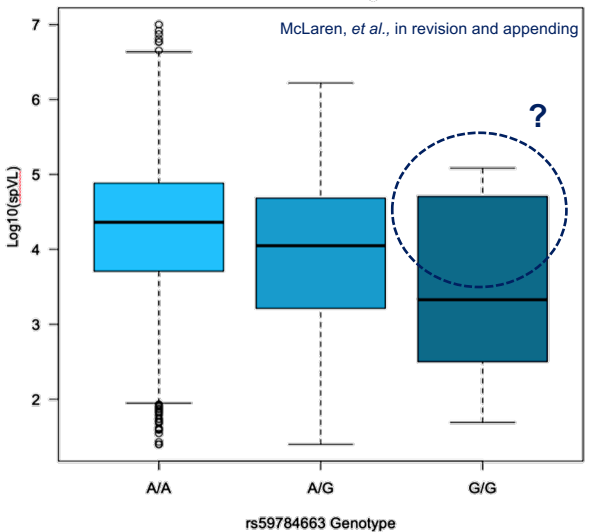
Linkage of HIV Escape Mutations to a Novel Host Genomic Locus Associated With Control of HIV Replication

Vanessa Schulz[1,2], Rupert Capina[2], Jeff Tuff[2], Lara Lewis[3], Joshua Kimani[4], Benjamin Chimukangara[3,5,6], Lyle R. McKinnon[1,3,4], Ayesha Kharsany[3], Paul J. McLaren[1,2]

[1]Department of Medical Microbiology & Infectious Diseases, University of Manitoba., [2]National HIV and Retrovirology Lab at the JC Wilt Infectious Diseases Research Centre., [3]Centre for the AIDS Programme of Research in South Africa., [4]Department of Medical Microbiology, University of Nairobi., [5]KwaZulu-Natal Research Innovation and Sequencing Platform, University of KwaZulu-Natal., [6]Department of Virology, University of KwaZulu-Natal



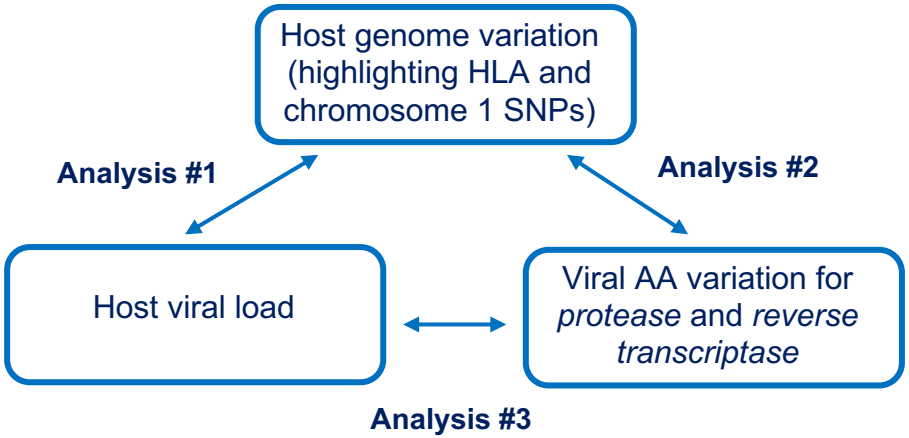
Our lab previously conducted a genome wide association study (GWAS)[1] in ~3,900 HIV⁺ individuals of African ancestry that identified a **novel locus on chromosome 1**. The **top associated variant rs59784663 (G)**, was associated with a ~0.3 log10 reduction in HIV set point viral load (spVL) and was downstream of the protein coding gene *CHD1L*. Association of rs59784663 and spVL is **unique to individuals of African ancestry**.

However, some individuals with the protective rs59784663 allele still experience high spVLs. Here, we hypothesized that *HIV develops escape mutations in response to host pressure from HLA and chromosome 1*

Methods

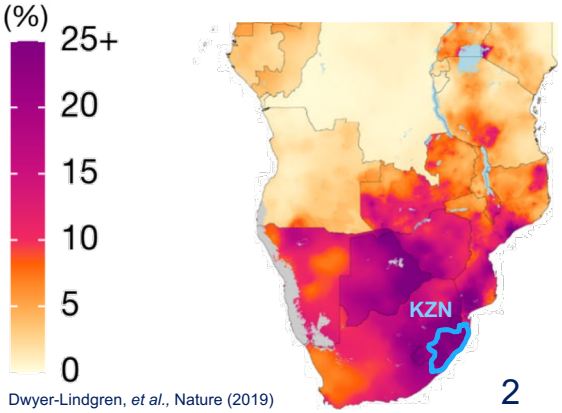
We performed 3 analyses in **HIV⁺ South African individuals (n=552)** part of the HIV Incidence Provincial Surveillance System (HIPSS)[2] with available **HIV sequence data (n=97)**[3] to identify amino acid (AA) variants in **HIV *protease* (PR)** and ***reverse transcriptase* (RT)** genes.

We investigated the **HLA region** on chromosome 6 and **near *CHD1L* on chromosome 1**. These SNPs were tested for association with AA variants HIV PR and RT to **identify escape mutations in response to host genomic pressure and to identify how these host-pathogen interactions affect VL.**



A summary of clinical data for the HIPSS cohort after QC filtering (n=552)	
Number of individuals with viral load data	508
Number of individuals no longer on ARVs (>1month)	54
Number of ARV naïve individuals	498
Mean log10 viral load	3.67
Mean CD4⁺ T cell count	641.7

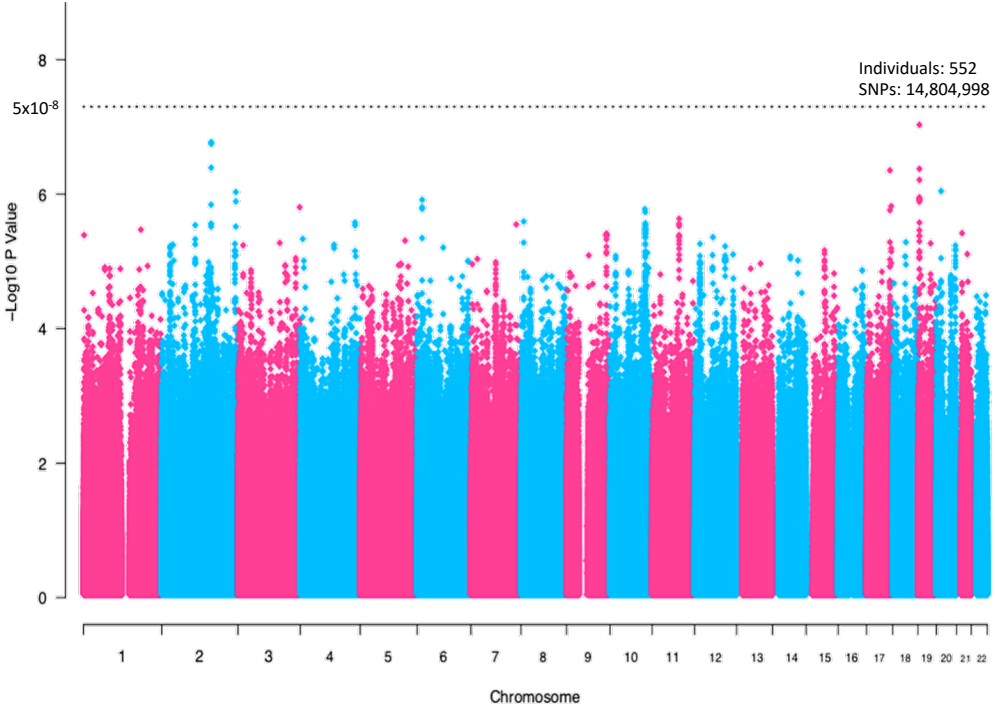
The HIPSS study was done in KwaZulu-Natal (KZN), South Africa. **South Africa has the highest burden of HIV worldwide and KwaZulu-Natal is the most affected province**[2,4].
The HIPSS cohort represents an understudied population at high-risk of acquiring HIV — an opportunity to study HIV co-evolution

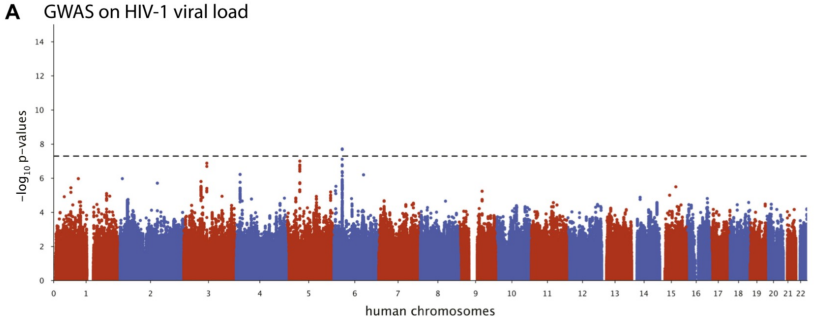


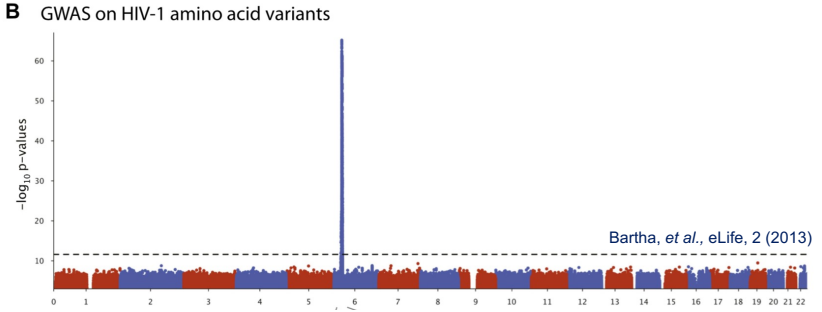
Dwyer-Lindgren, *et al.*, Nature (2019)

Host SNPs alone are not significantly associated with viral load

Analysis 1: A GWAS to identify host SNPs that are significantly ($p<5x10^{-8}$) associated with VL. After TOPMed imputation 14,807,459 SNPs were tested for association with VL in GEMMA using a univariate linear mixed model and likelihood ratio test. No significant associations were seen in this analysis.







Previously, Bartha, *et al.*, 2013[5] conducted genome-to-genome analyses on ~1,000 individuals of European ancestry.

They found that **host genome-to-viral genome analyses (B) produced stronger associations than host genome-to-viral load (A)**.

Moving into Analysis 2 we anticipated similar findings.

Host SNPs have stronger associations with viral variants than viral load

Analysis 2: Regional association analysis to identify host SNPs that are significantly associated with viral AA variants. Twenty 2-digit resolution HLA alleles and 4 chromosome 1 SNPs were tested for association with AA variants in HIV RT using logistic regression in PLINK. The significance threshold for HLA associations was $p<2.5x10^{-3}$ and the significance threshold for chromosome 1 associations was $1.25x10^{-2}$. Due to linkage disequilibrium between rs59784663 and rs73004025, they are listed together. The AA change for each codon is also listed.

A summary of significant associations between host alleles and viral AA variants for HIV RT

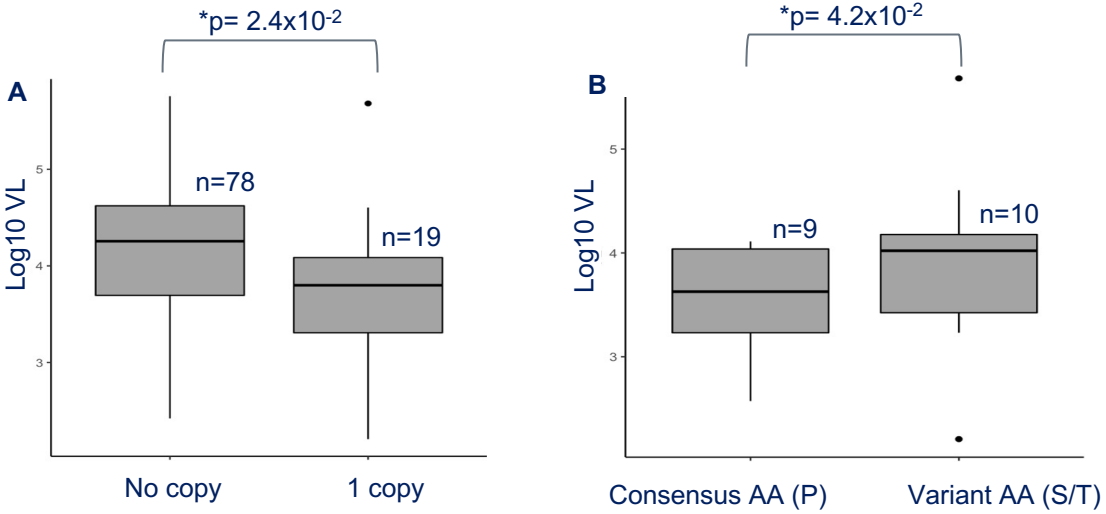
Host allele	Viral codon	AA change	p-value	OR
rs59784663/rs73004025	248	E248D	$9.9x10^{-3}$	5.19
HLAB*81	4	P4T, P4S	$1.5x10^{-5}$	20.18
HLAC*18	4	P4T, P4S	$1.4x10^{-3}$	9.12
HLAB*58	196	G196E, G196K	$1.0x10^{-3}$	8.09

Viral amino acid variants are not significantly associated with viral load

Analysis 3: To identify associations between viral AA variants and VL. Linear regression was done in PLINK with the significance threshold set at $p<1.72x10^{-3}$. There were no significant associations between VL and variant AA for PR and RT genes

Allele dosage effects on viral load suggest HIV develops escape mutations in response to host pressure

We investigated how the dosage of significant host alleles from analysis 2 affected VL. In addition, we wanted to see how viral AA variants in individuals with these alleles affected VL. Using a two-sample t-test we found that **VL was significantly lower (A), ~0.45 log10, in individuals that had 1 copy of the HLAB*81 allele** than those without a copy of the allele **($p=2.4x10^{-2}$)**.More notably, we found that **VL was significantly higher (B), ~0.37 log10, with a variant AA in codon 4 of RT** in individuals with the HLAB*81 allele **($p=4.2x10^{-2}$)**. This suggests that HIV has developed escape mutations in codon 4 of RT in response to HLAB*81 pressure.



Summary

- Host SNPs alone in the HIPSS cohort are not significantly associated with VL.
- Host SNPs in the HIPSS cohort have stronger associations with HIV AA variants in PR and RT than VL.
- Viral AA variants in RT and PR were not significantly associated with VL.
- 1 copy of the HLAB*81 allele results in lower VLs. Variable AA in RT codon 4 in an individual with HLAB*81 results in higher VL, suggesting viral escape.

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Contact: schulzv@myumanitoba.ca
McLaren Lab: https://mclarenlab.wixsite.com/mclaren-lab/about-the-team

Sources:

1. McLaren, *et al*., in revision and appending
2. Kharsany, *et al*., JAMA Network Open, 2 (2013).
3. Chimukangara, *et al*., AIDS Research and Human Retroviruses, 35 (2019).
4. Thami & Chimusa, Frontiers in Genetics, 10 (2019).
5. Bartha, *et al*., eLife, 2 (2013).