Signature of CD4 T Cells During Acute HIV-1 Infection is Predictive of Disease Progression

Dominic Paquin-Proulx[1,2], Bonnie M. Slike[1,2], Ningbo Jian[1,2], Boonrat Tassaneetrithep[1,2,3], Leigh Anne Eller[1,2], Gina Donofrio[1,2], Matthew Creegan[1,2], Sandhya Vassan[1,2], Julie A. Ake[1], Mary A. Marovich[1], Nelson L. Michael[4], Merlin L. Robb[1,2], Michael A. Eller[1,2], and Shelly J. Krebs[1] on behalf of the RV217 study team

[1]U.S. Military HIV Research Program, CIDR, Walter Reed Army Institute of Research, Silver Spring, MD; [2]U.S. Military HIV Research Program, Henry M. Jackson Foundation for the Advancement of Military Medicine, Bethesda, MD; [3]Center of Research Excellence in Immunoregulation, Research Department, Faculty of Medicine Siriraj Hospital, Mahidol University, Bangkok, Thailand, [4]Center for Infectious Disease Research, Walter Reed Army Institute of Research, Silver Spring, MD, USA

Abstract

Immune activation, a hallmark of chronic HIV infection, is a major contributor of HIV pathogenesis, even in the context of viral suppression by anti-retroviral therapy. The magnitude of immune activation during HIV infection is both established early and is more predictive of the rate of disease progression than plasma viral load. Elucidating the critical events in acute HIV infection (AHI) associated with disease progression will facilitate the design of new approaches to achieve HIV remission.

To investigate the dynamics of inflammation and immune activation during AHI, we utilized samples from 17 Thai and 26 East African participants in the RV217 AHI study[1]. All East Africans were female, and Thais mostly consisted of males and transgenders. Accordingly, the route of HIV acquisition was different between the study participants from East Africa and Thailand. In addition, each population consisted of different infecting subtypes. CRF01_AE was the infecting subtype in all but two Thai participants, while the subtypes in East Africa were a mixture of A, C, A/D, A/C, and A/C/D. The level of soluble and cellular markers of immune activation between East Africa and Thailand significantly differed between regions both before and during AHI. Similarly, variable signatures of immune activation were associated with peak and set point viral loads for each region. IP-10, IL-8 and IL-10 levels were associated with peak viral load in Africa but not in Thailand. Levels of IL-1β pre-infection in Thailand, but not in East Africa, were inversely associated with peak and set point viral loads. Combining datasets, higher levels of CCR5 and CD38 expression on CD4 T cells at 4.5 weeks post 1st HIV RNA positive test (around set point viral load) were predictive of reaching a CD4 count <350 independently of region or viral load.

Our results show that the frequency of CD4 T cells expressing CCR5 and CD38 at set point viral load is associated with faster disease progression independent of viral load. This suggests that CD4 T cells circulating with an activated phenotype associated with susceptibility to viral entry may be predictive of immune pathology.

Methods

We measured the levels of 26 soluble markers of inflammation inclusive of chemokines, cytokines, and markers of microbial translocation starting from pre-infection through AHI. The dynamics of CD4 and CD8 T cell activation during AHI were assessed using flow cytometry. The level and frequency of immune activation was used to predict a CD4 T cell count <350 using nonparametric rank-based testing, logistic regression, and cumulative incidence. The prediction capability of identified markers was also examined by Area Under the Receiver Operating Characteristic (AUROC) curve.

Results

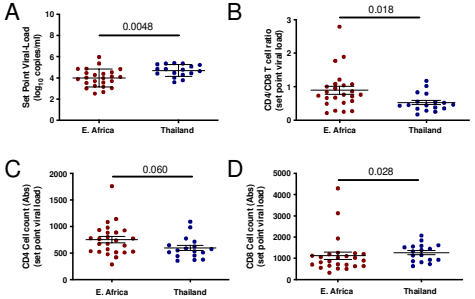

Figure 1. Differences in viral load, CD4, and CD8 T cell at set point viral load between participants from East Africa and Thailand. Participants from Thailand have a higher set point VL (A), as well as lower CD4 to CD8 T cell ratio (B), lower CD4 T cell absolute count (C), and higher CD8 T cell count (D). No differences were observed at peak viral load. * represent p values < 0.05 and ** represent p values < 0.01 (Mann-Whitney test), N=24 for East Africa and N=17 for Thailand.

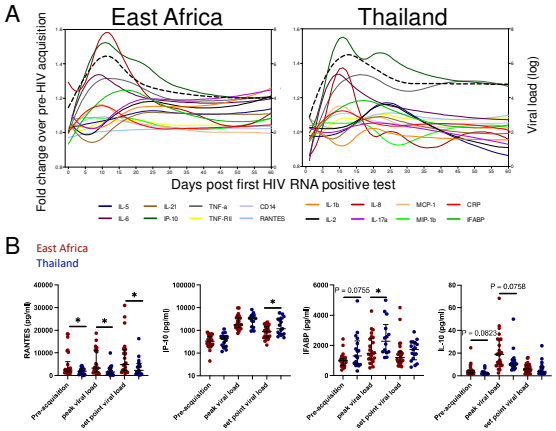

Figure 2. Dynamics of soluble markers during AHI. Spline model showing fold change from pre-HIV acquisition for each soluble marker for participants from East Africa (left) and Thailand (right) (A). Absolute values of RANTES, IFABP, IP-10, and IL-10 during AHI (B). * represent p values < 0.05 and ** represent p values < 0.01 (Mann-Whitney test), N=26 for East Africa and N=17 for Thailand.

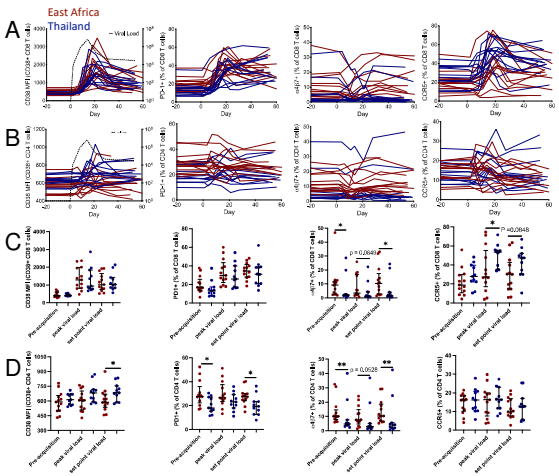

Figure 3. Dynamics of cellular activation during AHI. Dynamics of CD38 (far left, PD-1, center left, α4β7 (center right), and CCR5 (far right) on CD8 (A and C) and CD4 (B and D) T cells during AHI. * represent p values < 0.05 and ** represent p values < 0.01 (Mann-Whitney test), N=13 for East Africa and N=11 for Thailand. In panels A and B, the bold line represent the median for each group.

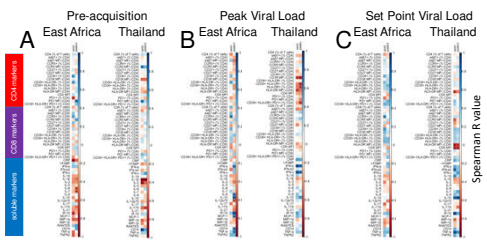

Figure 4. Spearman rank correlations of soluble factors and cellular immune activation at pre-acquisition (A), peak viral load (day 10 -18) (B), and early setpoint viral load (day 24-43) (C) with setpoint and peak viral load for participants from East Africa and Thailand. Stars indicates evidence of significance, where * represent p<0.05, and ** represent p<0.01.

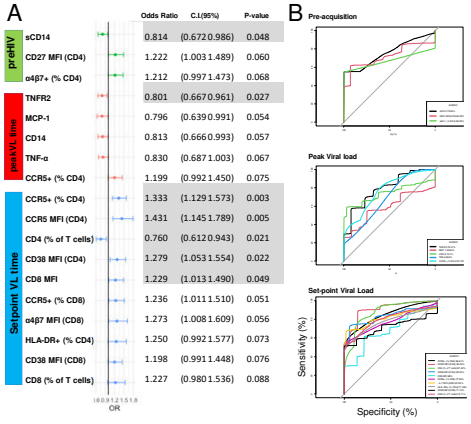

Figure 6. Markers with evidence of predicting status of reaching a CD4 count <350. Forest plot showing Odds ratio, 95% confidence interval, and p-values (A). Unit change for logistic regression is 1 standard deviation (SD) for cellular and soluble markers. Markers that are significant are highlighted in grey. Area under the receiver operating characteristic (AUROC) is shown for markers that are significant or trending at each time (B). All participants included, cohort difference and viral load is taken into consideration.

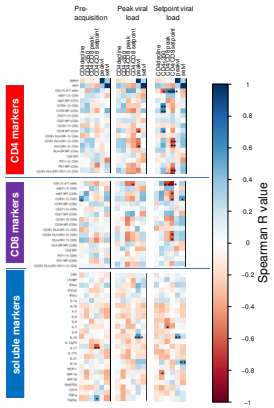

Figure 5. Spearman rank correlations of soluble factors (N=43) and cellular immune activation (N=24) with markers of HIV-1 disease progression at pre-infection, peak viral load (day 10 -18), and early setpoint viral load (day 24-43) when combining participants from East Africa and Thailand. Reaching CD4 count <350 was determined using all time points available prior to ART initiation with mean of 892 days of observation. Stars indicates evidence of significance, where * represent p<0.05, ** represent p<0.01, and *** represent p<0.001. Benjamini-Hochberg Procedure is applied for multiple testing correction to decrease False Discovery Rate (FDR).

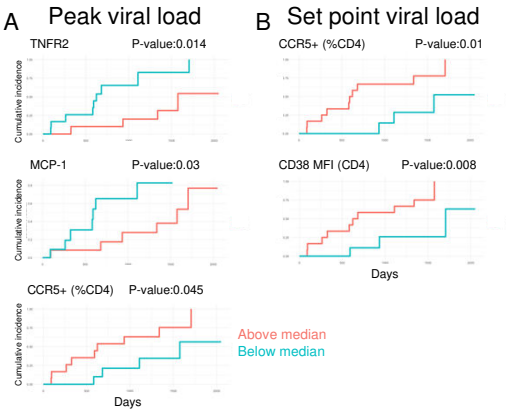

Figure 7. Cumulative incidence curves showing time to reaching a CD4 count <350 for participants with above or below the median level for each indicated marker at peak (A) and set point (B) viral load. All participants were included. P values are shown on each graph.

Conclusion

Through an extensive characterization of immune activation during AHI we have identified that different signatures are associated with peak or set point viral load in East Africa and Thailand. Differences in participants' demographics and viral subtype may have contributed to this phenomenon. Furthermore, our results suggest that that set point viral load is a critical time window that impacts HIV disease progression. In particular, CD4 T cells with a phenotype associated with susceptibility to HIV replication at set point viral load were correlated faster HIV disease progression.

Reference

1) Robb, M.L., et al. (2016). Prospective study of acute HIV-1 infection in adults in East Africa and Thailand. N. Engl.J. Med. 374, 2120–2130.

Material has been reviewed by the Walter Reed Army Institute of Research. There is no objection to its presentation and/or publication. The opinions or assertions contained herein are the private views of the author, and are not to be construed as official, or as reflecting true views of the Department of the Army or the Department of Defense. The investigators have adhered to the policies for protection of human subjects as prescribed in AR 70–25.