Unique and Common T-Cell Features Predicting Time-to-Rebound in Interventional and Non-Interventional ATI Trials 484

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Background

Immunological features predicting time-to-rebound during analytical treatment interruption (ATI) may differ between cohorts, and be impacted by whether or not therapeutic interventions were implemented. Here, we used CyTOF to identify shared and unique pre-ATI features of T cells associated with time-to-rebound in the non-interventional ACTG A5345 cohort and 3 interventional Danish cohorts.

Methods

A 40-parameter CyTOF T cell phenotyping panel was applied on pre-ATI blood specimens from 33 chronic-treated and 11 acute-treated individuals from the non-interventional ACTG A5345 cohort, and 3 chronic-treated Danish cohorts receiving therapy prior to ATI: CLEAR (Panobinostat intervention, n=9), TEACH (TLR9 agonist intervention, n=9), and REDUC (therapeutic vaccination + romidepsin intervention, n=16). Clustering analysis was performed to identify T cell phenotypes associated with time-to-rebound upon ATI. Label transfer analysis was performed to identify shared T-cell features between non-interventional and interventional ATI cohorts.

Results

Using cluster-resolution optimization, nine and five clusters were identified from the A5345 and the 3 collective Danish cohorts, respectively. Cluster A9 and A5 from A5345, and Cluster D4 from TEACH were all significantly ($p<0.01$) positively associated with longer time-to-rebound. Cluster A9 consisted of memory T cells expressing high levels of the T resident memory (Trm) marker CD103, and high levels of the homeostatic proliferation marker CD127 and the Th17 marker CCR6. As Cluster A9 consisted of both CD4+ and CD8+ T cells, we split it into subclusters A9a (CD4+) and A9b (CD8+). Cluster A9a uniquely expressed high levels of the co-stimulatory molecule CD28. By contrast, Cluster A9b expressed high levels of resident memory T cell markers CD49d and CD69. Cluster D4 from the Danish cohorts consisted of CD8+ T cells expressing high levels of pro-survival factor BIRC5 and CD28, and low levels of CD29. These cells also expressed high levels of CXCR4 and CCR7, suggesting homing to inflamed and lymphoid tissues. Cluster migration analysis identified cross-cohort equivalents of A5345 Cluster A5 and Danish Cluster D4. Overall, these results suggest that phenotypic features predicting longer time-to-rebound can be shared among non-interventional and interventional ATI cohorts.

Main findings

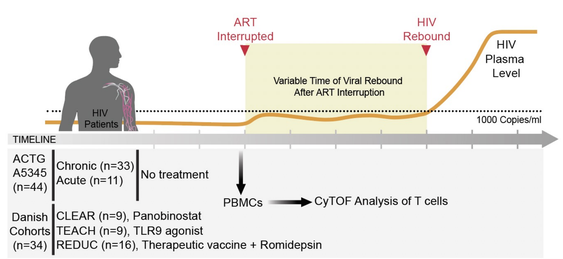
- ❖CyTOF clustering identified 9 subsets of T cells from non-interventional ACTG A5345 cohort
- ❖CyTOF clustering analysis identified 5 subsets of T cells from the 3 interventional Danish cohorts
- ❖Resident memory T cells were significantly associated with longer time-to-rebound in ACTG A5345
- ❖A subset of BIRC5+CXCR4+ CD8+ T cells were significantly associated with longer time-to-rebound in TEACH
- ❖Label transfer analysis revealed a shared subset of CD8+ T cells predicting longer time-to-rebound in both ACTG A5345 and TEACH cohorts

Future Directions

- ❖NK cell analysis from the same samples to identify NK features associated with rebound time
- ❖RNAseq analysis from the same samples to identify global immune features associated with rebound time

Study Design and Results

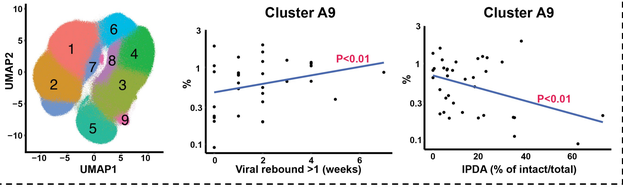
Schematic of the ACTG and Danish cohorts of PWH undergoing ATI



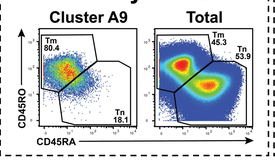
CyTOF panel for T cell phenotyping

Lineage Markers		Phenotyping Markers				
T Cells:	Differentiation State:	Activation State:	Homing Receptors:	Checkpoint Molecules:	Adhesion and integrins:	
CD3	CD45RO	CD69	CCR5	PD1	CD49d(α4)	
CD4	CD45RA	CD25	CCR6	TIGIT	CD103	
CD8	CD62L	CD28	CCR7	CTLA4	CD29(β1)	
B Cells:	CD57	CD30	CXCR4		α4β7	
CD19	CD127	HLADR	CXCR5			
	CD27	CD38				
	RORγt	ICOS				
	Tbet	OX40				
	CRTH2					
	CD73					
	Blimp1					
	CD7					

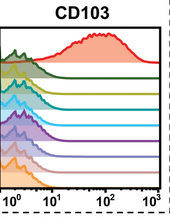
ACTG Cluster A9 is associated with longer time-to-rebound and lower intact provirus



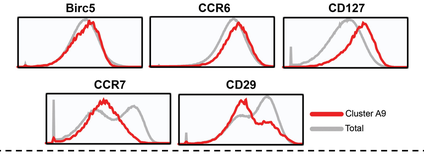
Cluster A9 cells are memory T cells



Cells in Cluster A9 uniquely express high levels of Trm marker CD103

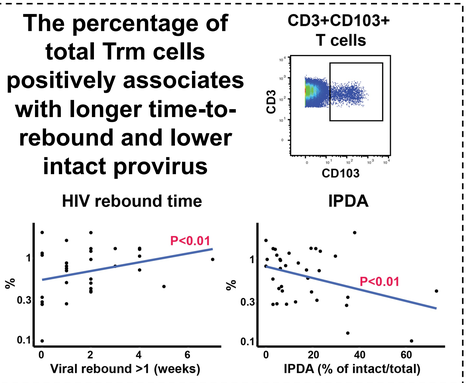


Cluster A9 cells express high levels of Birc5, CCR6 and CD127, and low levels of CCR7 and CD29

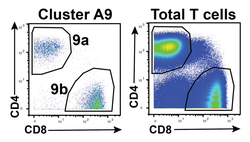


Results

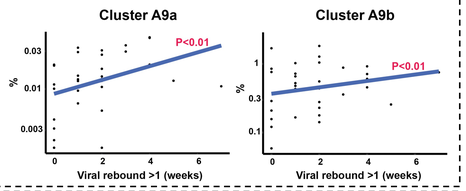
The percentage of total Trm cells positively associates with longer time-to-rebound and lower intact provirus



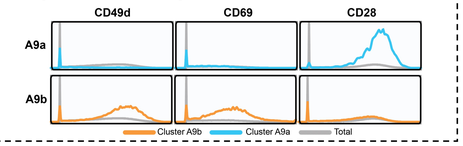
Cells in Cluster A9 include both CD4+ and CD8+ T cells



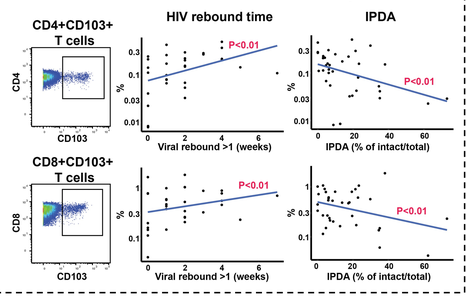
Clusters A9a and A9b are each independently associated with longer time-to-rebound



Unique marker expression in Clusters A9a/b

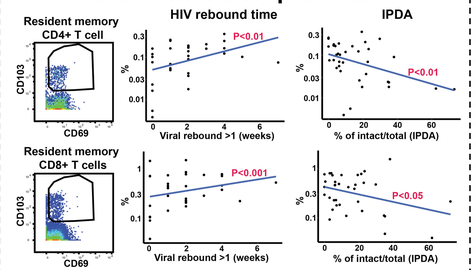


The percentages of CD4+ and CD8+ Trm cells are each independently associated with longer time-to-rebound and lower intact provirus

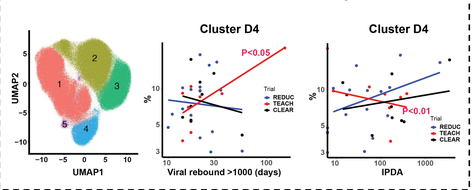


Results

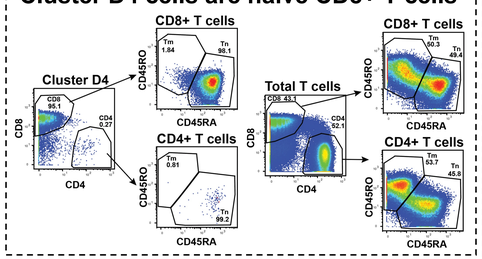
The percentages of CD103+CD69+ CD4+ and CD8+ Trm cells are each associated with longer time-to-rebound and lower intact provirus



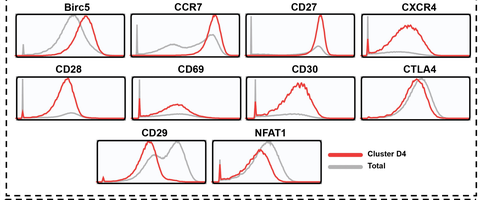
Danish cohort Cluster D4 is associated with longer time-to-rebound and lower intact provirus



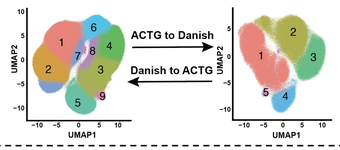
Cluster D4 cells are naïve CD8+ T cells



Marker expression profile of Cluster D4 cells

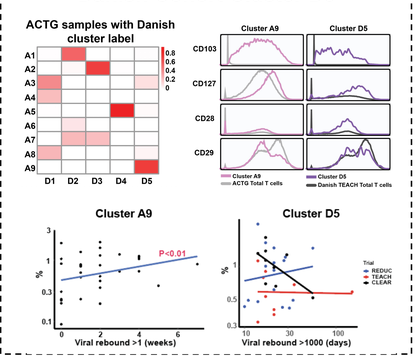


Label transfer analysis



Results

ACTG Cluster A9 corresponds to Danish Cohort Cluster D5



Danish Cohort Cluster D4 corresponds to ACTG Cluster A5

Conclusions

Our studies suggest CD4+ and CD8+ resident memory T cells (both CD103+ and CD69+CD103+) are associated with increased time-to-rebound in non-interventional treatment interruption. In the interventional TEACH cohort, a subset of CD8+ T cells with unique phenotypic features (including high expression of pro-survival factor BIRC5 and tissue-homing receptor CXCR4) are associated with increased rebound time. Similar cells also predict increased rebound time in ACTG A5345. These long-lived cells may be better able to home to lymphoid tissues during the early post-ART period and together with resident memory T cells slow viral rebound upon ATI.

Acknowledgements

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