HIV Research
UNIVERSITY OF TORONTO
Department of Medicine

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Mucosal-Associated Invariant T (MAIT) Cell Depletion and Exhaustion in HIV/HCV Co-infection.

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Presenter: Ali Fawaz

HIV and HCV Co-infection
November 18, 2013 – 10:55am

OHTN 2013 RESEARCH CONFERENCE
NOVEMBER 17-19, 2013

CHANGING THE COURSE OF THE HIV PREVENTION, ENGAGEMENT AND TREATMENT CASCADE

HIV/HCV Co-infection

- HCV targets the hepatocytes of the liver, and over time may cause cirrhosis.
- HIV/HCV co-infected patients progress more rapidly to end-stage liver disease, and compared to HCV mono-infection show:
  - Increased viral load.[1]
  - Higher rate of viral persistence[1]

- What is the immunological basis for this?

[1] Operskalski, E. a, & Kovacs, A. (2011). HIV/HCV co-infection: pathogenesis, clinical complications, treatment, and new therapeutic technologies. *Current HIV/AIDS reports*, *8*(1), 12–22.

Mucosal Associated Invariant T (MAIT) Cells

- Anti-microbial, innate-like T Cells
  - React against vitamin metabolites produced by bacteria and yeast
  - Characterized by expression of invariant TCR Va7.2, along with CD161 and IL-18R.
- Found in mucosal tissues, the liver, and peripheral blood.
- Constitute up to 10% of peripheral blood T cells, 40% liver T cells[2]
- Secrete IFNγ (anti-fibrogenic), TNFα and IL-17 (pro-fibrogenic), IL-22 (hepatoprotective)

[2] Young MH, Gapin L. 2013. Mucosal associated invariant T cells: don't forget your vitamins. Cell Research. 23: 460-462.

Question: Are MAIT Cells impaired in HIV/HCV?

- Due to their accumulation in the liver, and secretion of pro- and anti-fibrogenic cytokines, MAIT cells are of interest when examining liver disease progression.
- We wanted to know if:

1. **MAIT Cells are somehow impaired in HIV/HCV**
2. **This impairment could explain why liver disease progresses more rapidly in co-infected individuals.**

blood

2013 121: 1124-1135
Prepublished online December 13, 2012;
doi:10.1182/blood-2012-07-445429

Activation, exhaustion, and persistent decline of the antimicrobial MR1-restricted MAIT-cell population in chronic HIV-1 infection

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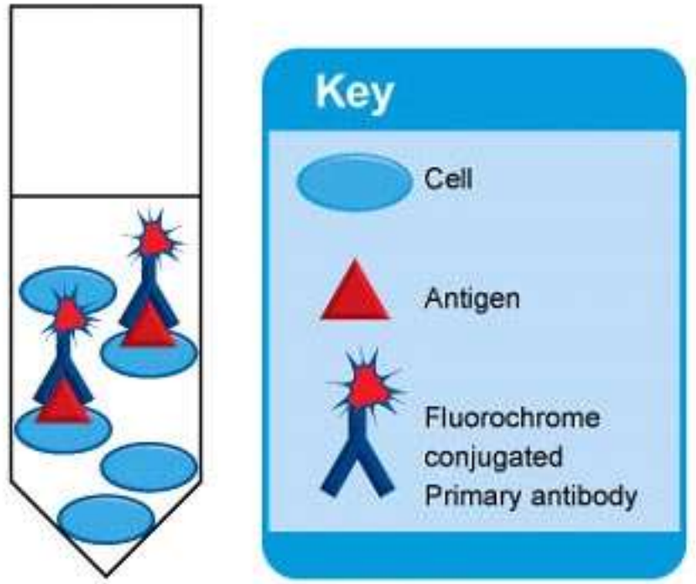
In HIV Mono-infection:

- MAIT cells are highly activated and exhausted (i.e. elevated Tim-3)
- Proportion of MAIT cells producing IFNγ, IL-17, and TNFα is lower.
- Reduced MAIT cell frequency in peripheral blood, static in rectal mucosa (potential recruitment?)
- Accumulation of less functional, CD161- Vα7.2+ MAIT cells as infection progressed.

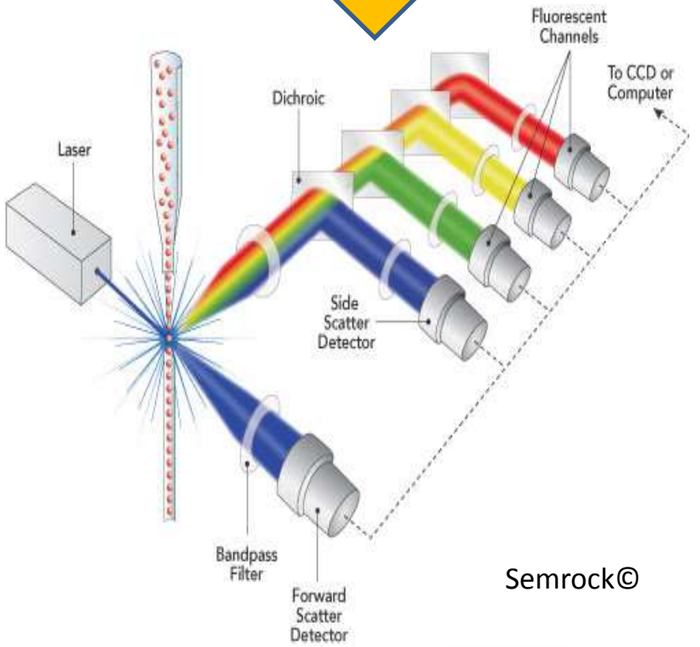
Methods: Flow Cytometry

- Peripheral blood mononuclear cells (PBMCs) from uninfected, chronic HIV+, HCV+, and HCV+/HIV+ patients were stained with fluorescent antibodies.
- Analyzed using flow cytometry, which allowed for:

1. Identification of MAIT Cells in a mixed PBMC population
2. Assessment of functional capacity.



Abcam®



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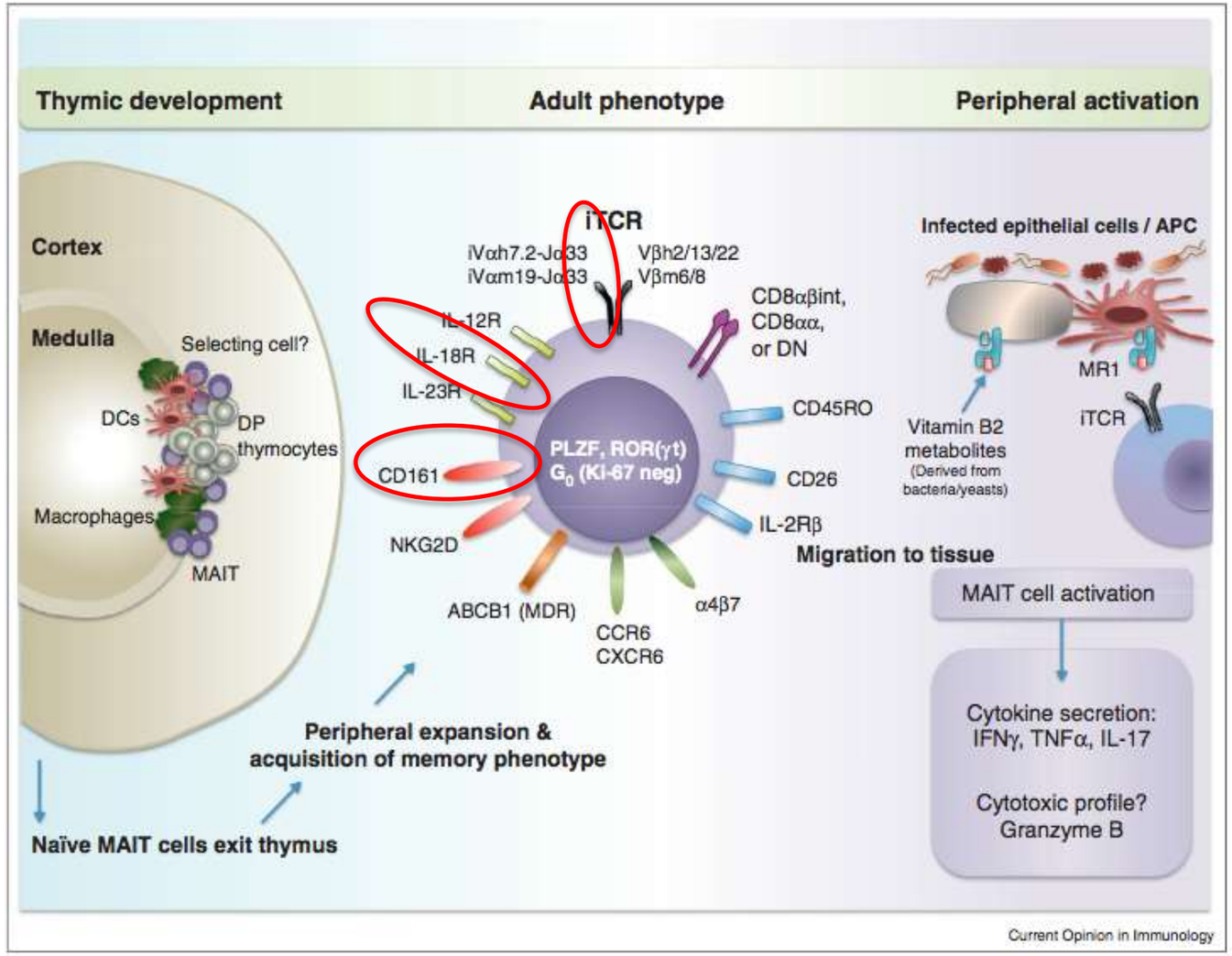
Flow Cytometry: Staining Panel

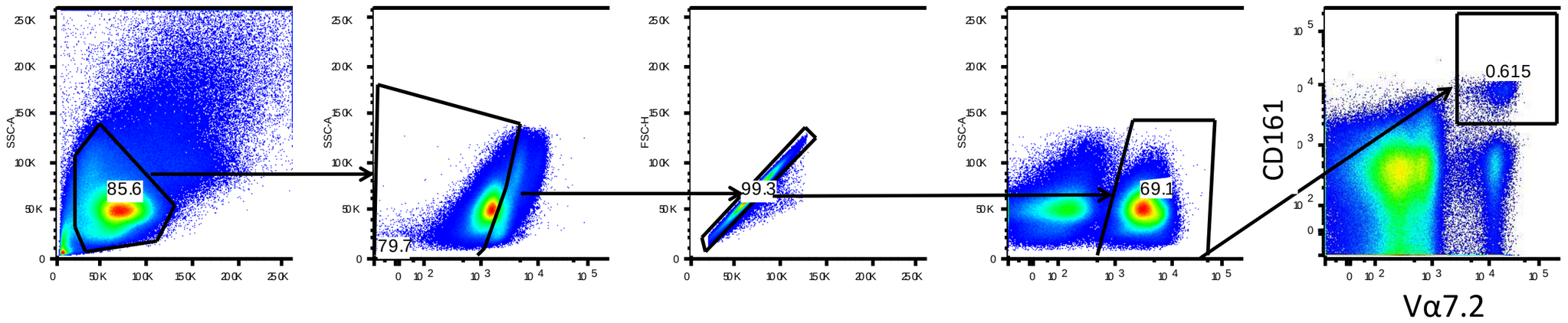
Marker	Function
CD3	TCR Component
Vα7.2	Specific to MAIT TCR
CD161	IL-17 Producing Cell Marker. Identifies MAIT Cells
PD-1	Exhaustion Marker
Tim-3	Exhaustion Marker
IFNγ	Anti-fibrogenic cytokine, marker of functional capacity.

Figure 1



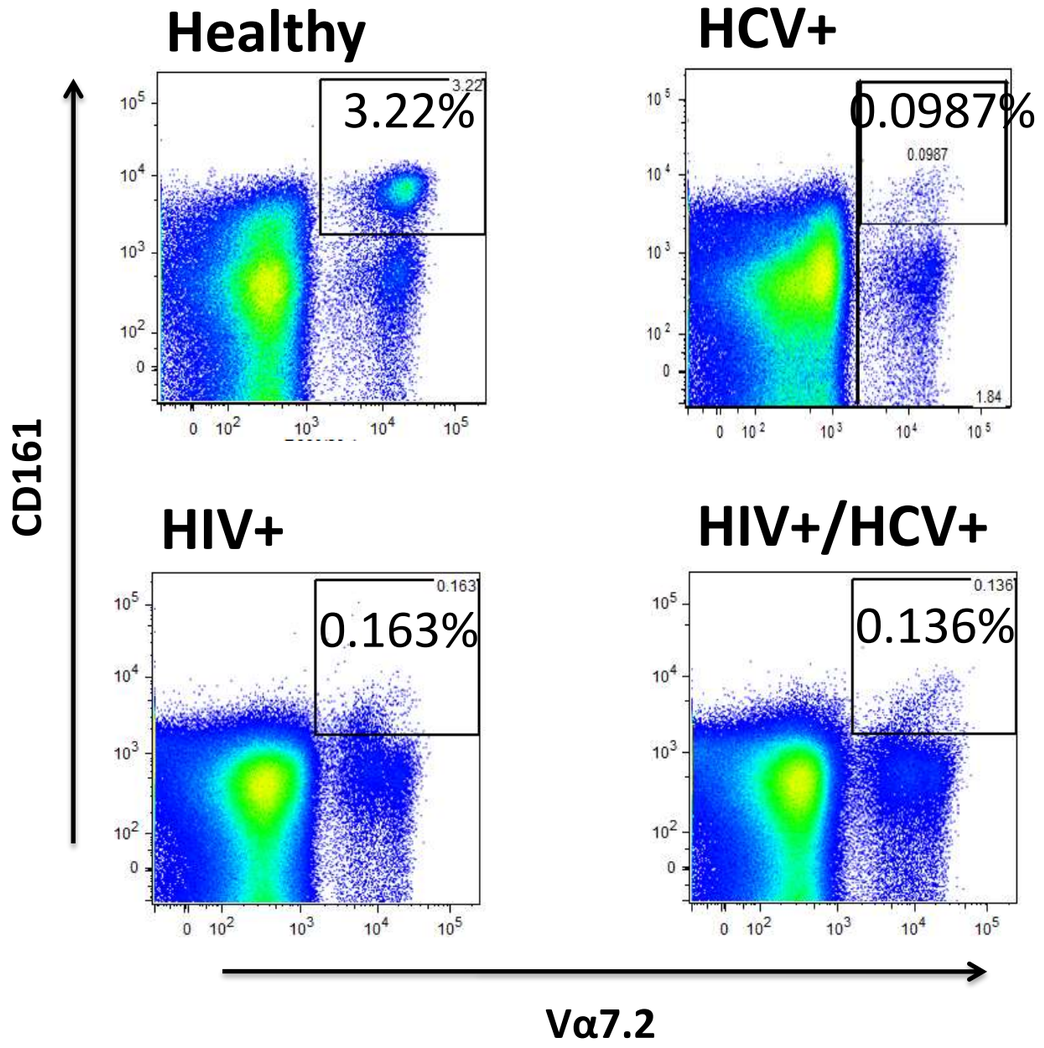
Development, phenotype, specificity and effector activities of MAIT cells.

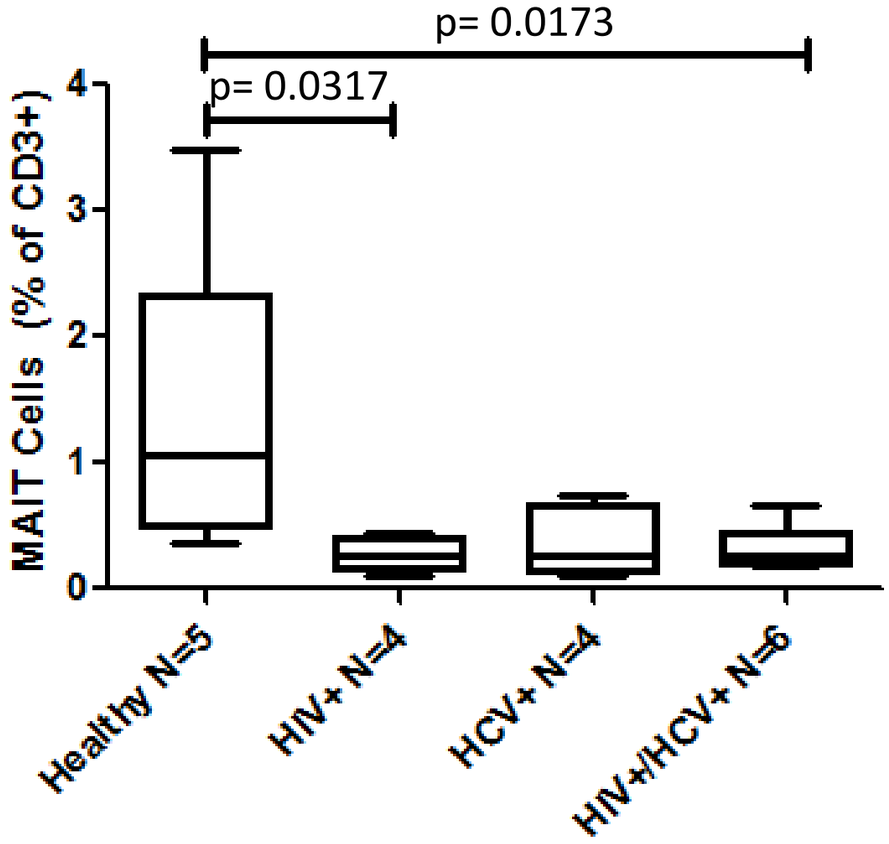
Gating Strategy



Lymphocytes **Live Cells** **Singlets** **CD3 +** **MAIT Cells**

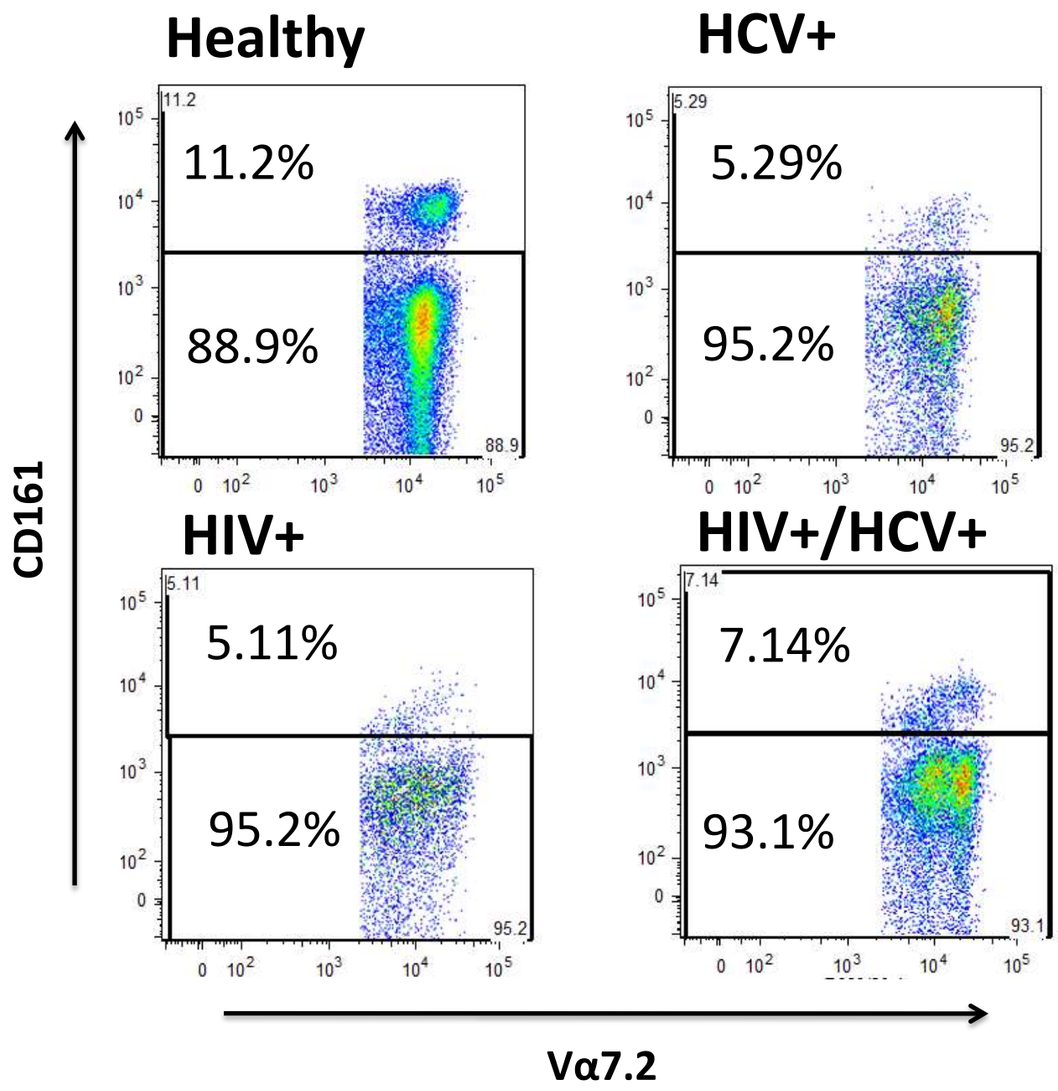
Depletion of MAIT Cells in Peripheral Blood of HIV, and HCV/HIV Patients

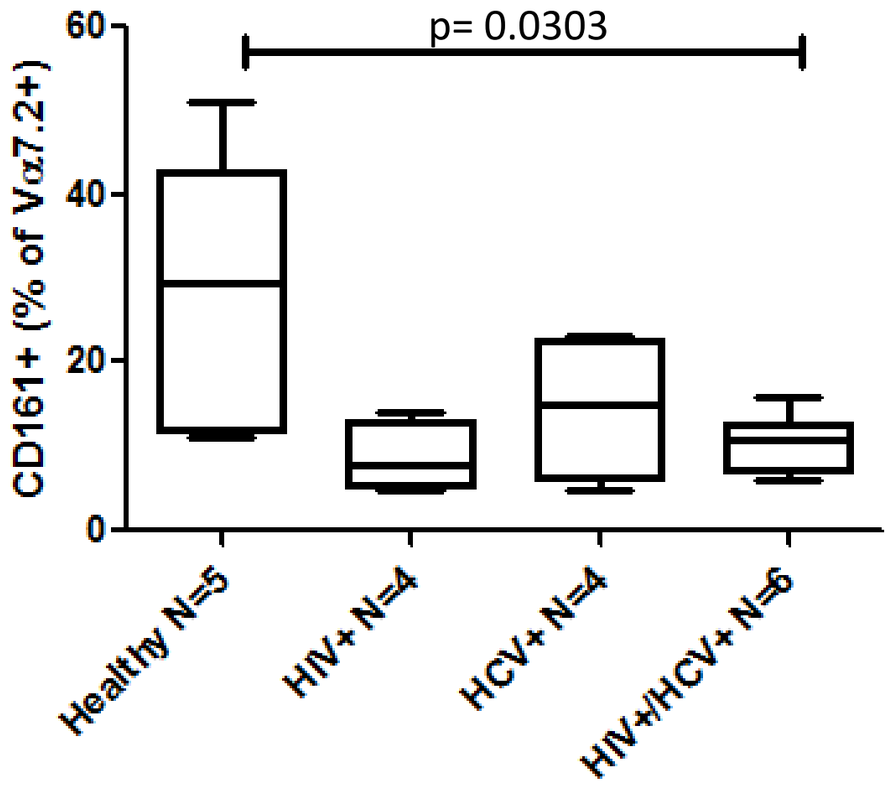




Two-tailed Mann-Whitney t test (95% confidence interval) was used for statistical analysis.

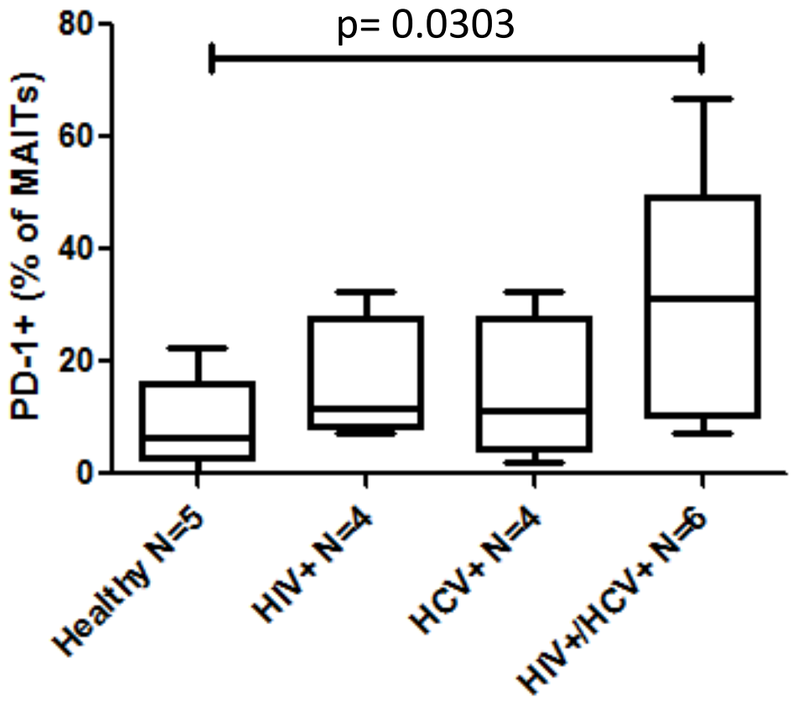
Depletion of CD161+/Vα7.2+ Subset in HIV/HCV



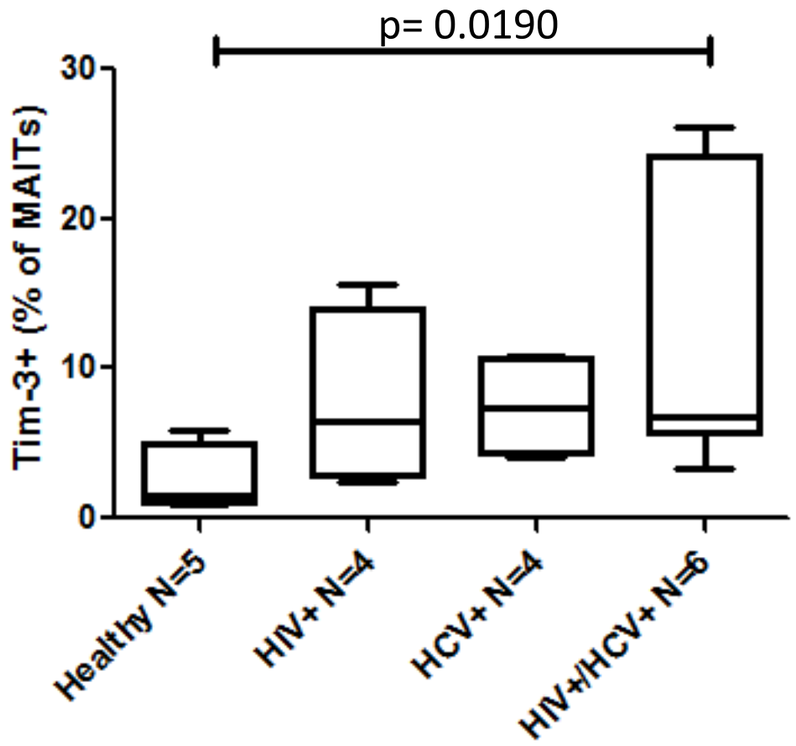


MAIT Cells Show Exhaustion Phenotypes in HIV/HCV

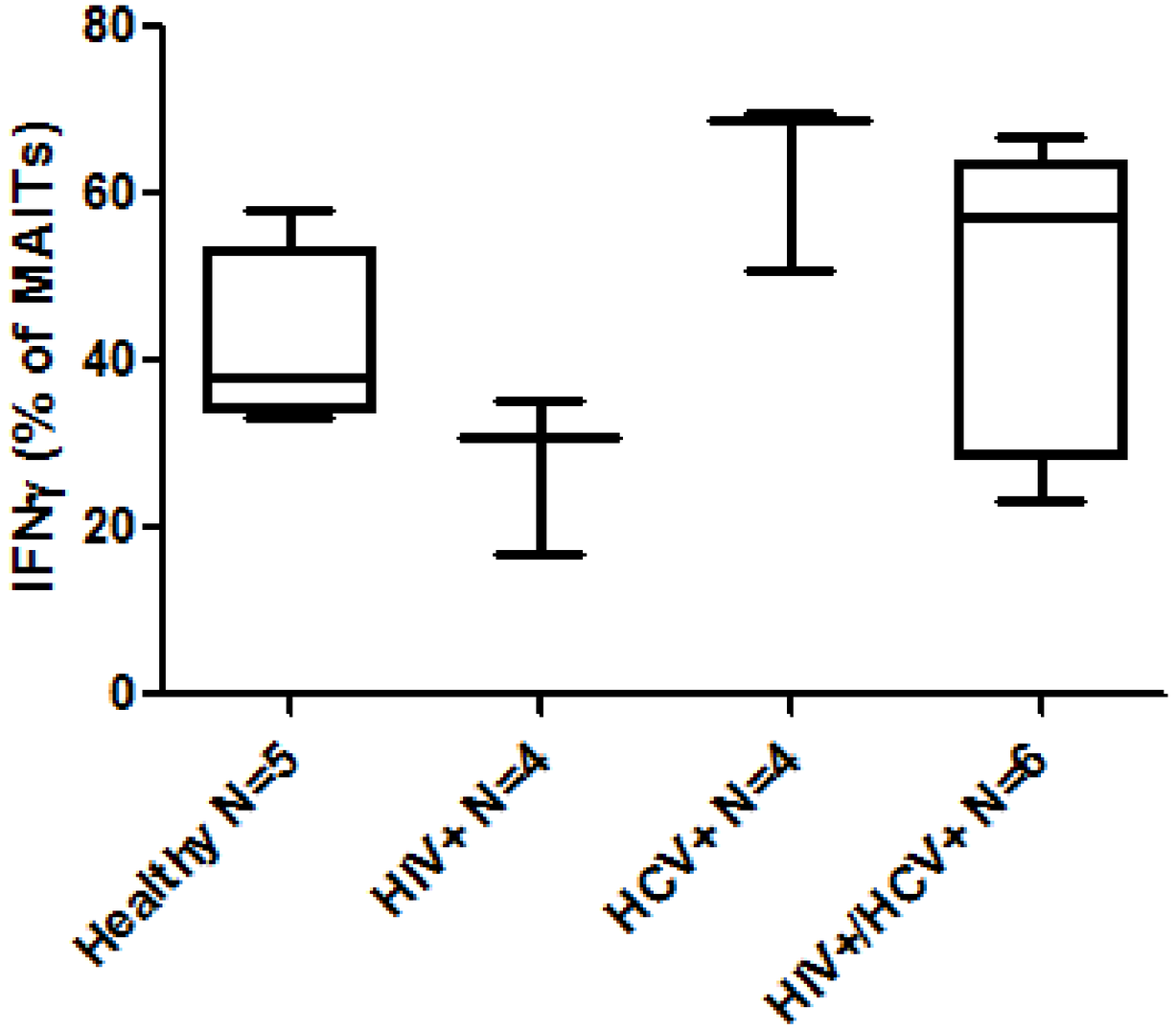
PD-1+ MAIT Cells:



Tim-3+ MAIT Cells



MAIT Cell Production of IFNγ with PMA/Ionomycin Stimulation



What Do These Findings Tell Us?

Going back to the original questions:

Are MAIT Cells impaired in HIV/HCV?:

- They are depleted in the peripheral blood.
- There is a significant decline in the proportion of functional (Vα7.2+, CD161+) MAIT Cells in HIV/HCV.
- Greater proportion of MAITs expressing the exhaustion markers PD-1 and Tim-3 in HIV/HCV.

Can this impairment explain the more rapid progression of liver disease in HIV/HCV?

- Difficult to make any conclusions at this point.
- Will become more clear as their functional capacity is more comprehensively characterized.

Future Directions

- **Assess MAIT cell phenotypes in the liver.**
- Re-assess IFNγ production using a more physiological stimulus, such as *E.coli*.
- Expand our functional characterization to the remaining cytokines.
  - Are there compounding negative effects? (ex. Lower IFNγ production together with higher IL-17)

Acknowledgements

University of Toronto HIV Group

Dr. Mario Ostrowski

Dr. Sonya MacParland | **Saleh Fadel** | Hampavi Sivanesan | Wade Coutinho

Ardalan Bozorgzad | Shariq Mujib | Dr. Jun Liu | Dr. Nur-ur-Rahman

Elizabeth Yue | Jordan Schwartz | Matthew Haaland | Kiera Clayton

Wei Zhan | Matthew Douglas-Vail | Yu Li

MacDonald Lab | **Kaul Lab**

Maple Leaf Medical Clinic

Colin Kovacs

National CIHR Research Training Program in Hepatitis C | Subvention nationale de formation des IRSC en hépatite C

CIHR IRSC
Canadian Institutes of Health Research | Instituts de recherche en santé du Canada

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