Polyfunctional SARS-CoV-2-specific T cells persist in tissue of COVID-19 convalescents

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Introduction

- T cells play an essential role in SARS-CoV-2 immunity, including in defense against severe COVID-19 (Chen, 2020; Neidleman, 2022, 2021; Moss, 2022).
- However, most studies analyzing SARS-CoV-2-specific T cells have been limited to analysis of blood.
- The role of T cells in SARS-CoV-2 immunity in pregnant women, which are at disproportionately higher risk of severe COVID-19 (Male, 2022), is poorly understood.

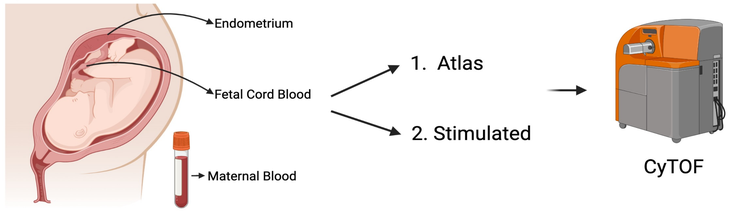
Study Questions

1. What are the frequencies of SARS-CoV-2-specific T cells in the endometrium, maternal blood, and fetal cord blood of COVID-19 convalescent pregnant women?
2. Are SARS-CoV-2-specific T cells from endometrium vs. blood phenotypically distinct? If so, what phenotypic features distinguish these cells?
3. What effector functions do SARS-CoV-2-specific T cells from endometrium and blood exhibit?

Experimental Design

- Endometrial tissue, maternal blood, and fetal cord blood were collected from convalescent pregnant women (n=12) at the time of delivery
- Infection times ranged from 3 days to 5 months before specimen collection
- Immune cells were isolated and split into two fractions – 1) Unstimulated "Atlas" 2) SARS-CoV-2 peptide + costimulatory antibodies (anti-CD49d and CD28) "Stimulated"
- Both fractions were barcoded, stained with lineage and phenotyping markers, and analyzed by CyTOF
- A sequential gating strategy was used to identify CD4+ and CD8+ T cells
- High-dimensional analysis tools were then used to characterize SARS-CoV-2-specific T cells in all three sites

Specimen collection at term:



CyTOF Panel

Lineage Markers

T Cells:
CD3
CD4
CD8

T Cell Subsets:
CD45RA
CD45RO
CD127
CD27 CD57
CD62L TCF1
Foxp3 CD95

Phenotyping Markers

Activation State:
ICOS
CD29
OX40
CD28
CD69
CD38
CD25
HLADR
4-1BB
Ki-67

Exhaustion/ Activation Markers:
PD1
TIGIT
CTLA4

Homing Receptors:
CCR5
CCR6
CCR7
CXCR4
CXCR5

Degranulation:
LAMP1
Granzyme B
Perforin

Cytokines
IFNγ
TNFα
IL2
MIP1β
IL4
IL21

References

- Chen et al. Clinical and immunological features of severe and moderate coronavirus disease 2019. J Clin Invest 2020; 130(5): 2620–2629.
- Neidleman et al. SARS-CoV-2-Specific T Cells Exhibit Phenotypic Features of Helper Function, Lack of Terminal Differentiation, and High Proliferation Potential. Cell Rep Med 2020; 1(6): 100081.
- Neidleman et al. Distinctive features of SARS-CoV-2-specific T cells predict recovery from severe COVID-19. Cell Rep 2021; 36(3):109414.
- Moss. The T cell immune response against SARS-CoV-2. Nat Immunol 2022; 23(2):186-193.
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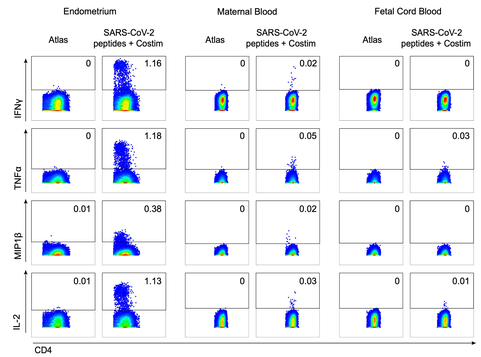
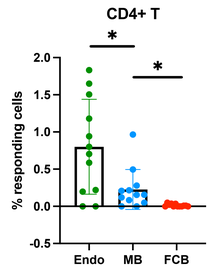
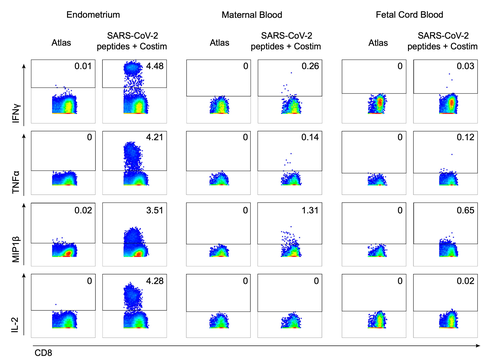
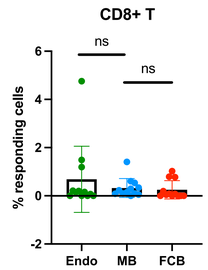
Results

The pregnant endometrium is highly enriched for SARS-CoV-2-specific T cells 5 months after infection

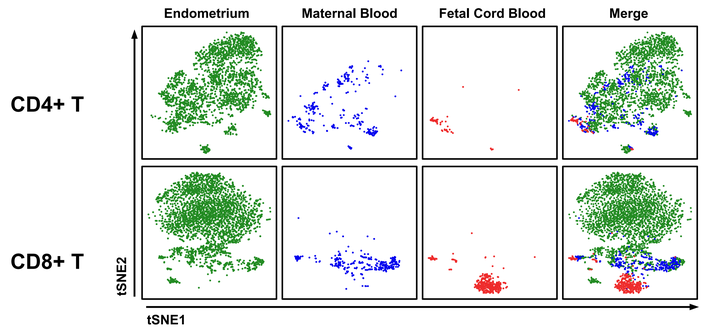




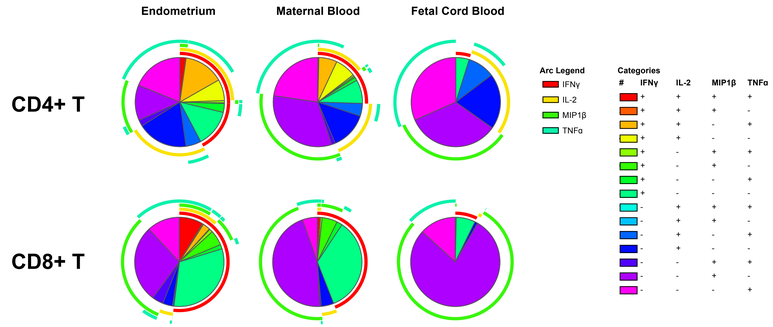




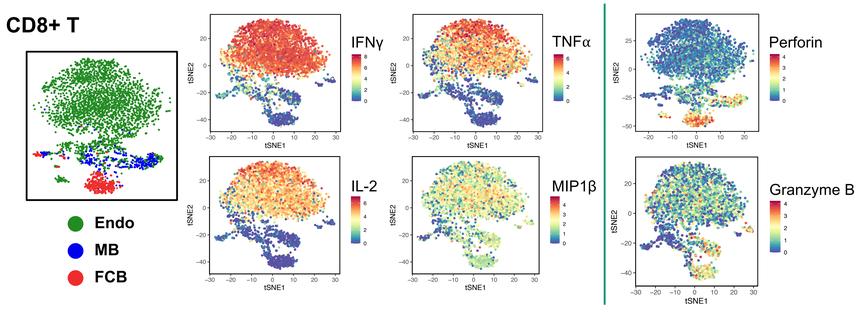
SARS-CoV-2-specific T cells from the endometrium are phenotypically distinct from those in blood



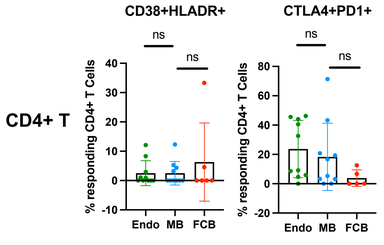
SARS-CoV-2-specific T cells from the endometrium are highly polyfunctional

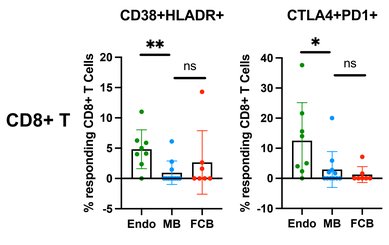


Endometrial SARS-CoV-2-specific CD8+ T cells produce high levels of cytokines while those from blood exhibit more cytolytic features



A subset of activated SARS-CoV-2-specific CD8+ T cells is enriched in the endometrium as compared to blood





Summary and Conclusions

- SARS-CoV-2-specific CD4+ and CD8+ T cells are enriched in the endometrium of COVID-19 convalescent pregnant women relative to frequencies in blood.
- These endometrial SARS-CoV-2-specific T cells can persist for up to 5 months post-infection.
- Endometrial SARS-CoV-2-specific T cells are highly polyfunctional, and are comprised of a higher proportion of quadra- and tri-functional responding cells.
- These effector cells preferentially produce high levels of multiple effector cytokines (IFNγ, TNFα, IL2, and/or MIP1β) in response to cognate peptide encounter.
- Blood-derived SARS-CoV-2-specific CD8+ T cells preferentially produce cytolytic effectors.
- A subset of activated endometrial SARS-CoV-2-specific CD8+ T cells co-expressing CD38 and HLADR, or CTLA4 and PD1, is enriched in endometrium as compared to blood. This enrichment was not observed in the CD4 compartment.
- The high cytokine but low cytolytic features of endometrial SARS-CoV-2-specific CD8+ T cells may be important in maintaining a tolerant yet anti-viral state in the decidua.

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