Monocyte Exposure to HBV Antigens Leads to Activation of HBV-specific CD4 T Cells.

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Hepatitis B Virus

- Hepatitis B virus infection
  - 250 million people chronically infected worldwide
    - vs. 150 mil HCV or 40 mil HIV
  - 300,000 patients in Canada
  - > 700,000 deaths/year worldwide

- Primary risk factor for liver cancer
  - >50% HBV-related
  - 2nd leading cause of cancer deaths

- Antiviral therapy rarely eliminates HBV
  - Nucleoside analogues
    - Block intermediate replication step
    - Does not eliminate viral template
  - IFN-alpha
    - Limited use due to side effects
Post-infection Clearance of HBV Requires T cells

**Acute**

Multi-specific T cell response + anti-core/anti-surface Abs
1. CD4 T helper cells required to support CD8 T cells & B cells
2. CD8 T cells mediate clearance of infected cells
3. B cells produce anti-HBs to clear circulating virus

**Chronic**

T cells are exhausted in chronic HBV patients
1. HBV-specific T cells are prone to apoptosis
2. Co-express inhibitory receptors – PD-1, CTLA-4, Tim-3

Mechanism driving T cell exhaustion is not well characterized

Hepatitis B Virus Immune Evasion/Exhaustion

- **High Antigen burden**
  - Small (3.2kb) DNA virus that infects hepatocytes
    - Can sustain up to $10^{10}$ HBV DNA copies/ml serum in chronic patients
    - Viral antigen can reach 1 mg/ml in serum

- Ex vivo sorted HBV patient APCs

- Monocytes internalize and retain HBsAg in the blood
- Monocytes can present antigen to CD4 T cells

- Gehring et al, J Clin Invest. 2013;123(9):3766-76
Hypothesis: Monocytes in the blood of chronic HBV patients continually present antigen to HBV-specific CD4 T cells, leading to T cell exhaustion.

Aim: Characterize the ability of blood monocytes to present HBsAg to HBV-specific T cells to better understand drivers of T cells exhaustion in chronic HBV infection.

Cloning & Characterization of HBsAg-specific CD4 T cell Clone

- CD4 T cell clones are HLA-DR11 restricted and HBs180 peptide specific.
Recombinant HBsAg is Internalized, Processed and Presented to CD4 T cell clones

Mammalian-derived HBsAg is Internalized, Processed and Presented to CD4 T cells

Mammalian cell line derived HBsAg activates T cells but to a lesser extent than yeast derived HBsAg
Different glycosylation, different lipid profiles
HBsAg in the Serum of Chronic HBV Patients is Internalized, Processed and Presented to CD4 T cells

Summary

• CD4 T cell clones are HBs Antigen specific and HLA-DR11 restricted.

• Monocytes present rHBsAg and mammalian cell line derived HBsAg antigen to CD4 T cell clones.

• HBsAg in the serum of Chronic HBV Patients is Internalized, Processed and Presented to CD4 T cells.

Future Directions

• Staining of monocytes to measure the frequency of HBsAg⁺ve monocytes in correlation to % IFN-γ⁺ve T cells.

• Confirm in vitro data using HLA-matched CD14 monocytes isolated ex vivo from different cohorts Chronic HBV patients.
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