EFFECTS OF IMMUNE CHECKPOINT THERAPY ON LATENT HIV IN PEOPLE WITH HIV AND MALIGNANCY

Jillian SY Lau1, James H McMahon1, Ajantha Solomon1, Chris YH Chiu2, Celine Gubser2, Ashanti Dantanarayana3, Socheata Chea4, Surekha Tennakoon2, Jennifer M Zerbato2, Jill Garlick1, Vincent Morcilla3, Sarah Palmer2, Sharon R Lewin1,2, Thomas Rasmussen1

1) Department of Infectious Diseases, Alfred Hospital and Monash University, Melbourne, Australia; 2) The Peter Doherty Institute for Infection and Immunity, The University of Melbourne and Royal Melbourne Hospital, Melbourne, Australia; 3) The Westmead Institute for Medical Research, University of Sydney, Westmead, Australia

Contact: jillian.lau@monash.edu @JillianLau1

Background
• Antibodies that block PD-1 and CTLA-4 demonstrate therapeutic efficacy in cancer by enhancing tumour-directed T cell responses.
• HIV persistence on antiretroviral therapy (ART) occurs preferentially in cells expressing immune checkpoints, including PD-1 and CTLA-4.
• PD-1 expression is associated with decreased function of antigen-specific T cells.1-3
• Immune checkpoint blockade (ICB) may impact HIV persistence on ART by enhancing HIV-specific CD8+ T cell responses and/or reversing HIV latency.4-7
• Here, we report on the effect of ICB on HIV latency and HIV-specific immunity in 3 PLHIV on ART who received ICB therapy for cancer.

Methods
• Samples obtained during the first 4 cycles of ICB (Fig 1).
• Virological assessments performed on blood CD4+ T cells:
  • Cell associated (CA) unspliced (US) HIV RNA
  • Cell associated (CA) HIV DNA
  • Tat/tat Induced Limiting Dilution Assay (TILDA)
• Plasma HIV RNA using a single copy assay (SCA)
• Flow cytometry was used to assess:
  • Frequency of Gag-specific CD4+ and CD8+ T cells positive for IFN-γ, TNF-α, or CD107a by intracellular cytokine staining (ICS).

Results
• Demographic and clinical data is summarised in Table 1. CA-US RNA increased following each infusion in all 3 participants. (Fig 2A, B).
• No consistent change in HIV-DNA was seen (Fig 2C).
• In Participant (P3) plasma VL increased from 4c/mL at baseline to 16c/mL on ICB and a reduction was seen in both HIV DNA and inducible MS RNA as measured by TILDA over 4 cycles of ICB (Fig 2D).
• P2 demonstrated an increase in the frequency of HIV- specific effector memory CD8+ T cells positive for IFN-γ, TNF-α, and CD107a by intracellular cytokine staining (ICS).

Conclusions
• Increases in HIV transcription were observed on ART in all 3 participants following each cycle of either anti-PD-L1 or combination anti-PD-1/anti-CTLA-4.
• Some participants (P3), combination anti-PD1/anti-CTLA-4 was associated with a reduction in HIV DNA and inducible virus.
• Combination anti-PD1/anti CTLA-4 was associated with increased HIV-specific immune responses, but only in one participant (P2).
• Our results highlight that ICB can activate latent HIV and increase HIV-specific immune responses, but with variation between individuals.

References

Acknowledgements
This work was funded by grants from the Australian NHMRC and ACH2. The authors would like to thank the HIV positive participants of this study.

Table 1. Participant demographics and clinical results.
<table>
<thead>
<tr>
<th></th>
<th>P1</th>
<th>P2</th>
<th>P3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age/sex</td>
<td>76/m</td>
<td>56/m</td>
<td>68/m</td>
</tr>
<tr>
<td>Malignancy</td>
<td>Merkel Cell Carcinoma</td>
<td>Metastatic Melanoma</td>
<td>Metastatic Melanoma</td>
</tr>
<tr>
<td>ICB</td>
<td>Avelumab (anti-PD-L1) 10mg/kg 2 weekly</td>
<td>Nivolumab (anti-PD-1) 3mg/kg + Ipilimumab (anti-CTLA-4) 1mg/kg 3 weekly</td>
<td>Nivolumab 3mg/kg + Ipilimumab 1mg/kg 3 weekly</td>
</tr>
<tr>
<td>HIV Diagnosis</td>
<td>HIV VL at BL</td>
<td>2001</td>
<td>2007</td>
</tr>
<tr>
<td>ART</td>
<td>TDF/3TC/EFV</td>
<td>TDF/3TC/EFV</td>
<td>TDF/3TC/EFV</td>
</tr>
<tr>
<td>CD4 nadir</td>
<td>53 cells/uL</td>
<td>210 cells/uL</td>
<td>53 cells/uL</td>
</tr>
<tr>
<td>Baseline CD4</td>
<td>323 cells/uL</td>
<td>468 cells/uL</td>
<td>265 cells/uL</td>
</tr>
<tr>
<td>Progress</td>
<td>No irAE</td>
<td>Partial response</td>
<td>Died before study completion</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Died after study completion</td>
</tr>
<tr>
<td></td>
<td>No irAE</td>
<td>Disease progression</td>
<td>Disease responsive</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Maintenance nivolumab</td>
</tr>
</tbody>
</table>

Table 1. Participant demographics and clinical results.
- **P1**: Participant, VL: viral load; BL: baseline; TDF, tenofovir disoproxyl fumarate; 3TC, lamivudine; FTC, emtricitabine; EFV, efavirenz; EVG, elvitegravir; COBI, cobicistat; TAF, tenofovir alafenamide; irAE, immune related adverse event.

**Fig 1. Clinical Trial design** "EOS: end of study"

**Methods**
- **Samples obtained during the first 4 cycles of ICB (Fig 1).**
- **Virological assessments performed on blood CD4+ T cells:**
  - Cell associated (CA) unspliced (US) HIV RNA
  - Cell associated (CA) HIV DNA
  - Tat/tat Induced Limiting Dilution Assay (TILDA)
- **Plasma HIV RNA using a single copy assay (SCA)**
- **Flow cytometry was used to assess:**
  - Frequency of Gag-specific CD4+ and CD8+ T cells positive for IFN-γ, TNF-α, or CD107a by intracellular cytokine staining (ICS)

**Results**
- **Demographic and clinical data is summarised in Table 1.** CA-US RNA increased following each infusion in all 3 participants. (Fig 2A, B).
- **No consistent change in HIV-DNA was seen (Fig 2C).**
- **In Participant (P3) plasma VL increased from 4c/mL at baseline to 16c/mL on ICB and a reduction was seen in both HIV DNA and inducible MS RNA as measured by TILDA over 4 cycles of ICB (Fig 2D).**
- **P2 demonstrated an increase in the frequency of HIV- specific effector memory CD8+ T cells positive for IFN-γ, TNF-α, and CD107a (Fig 3A, B) in response to Gag stimulation.**

**Conclusions**
- **Increases in HIV transcription were observed on ART in all 3 participants following each cycle of either anti-PD-L1 or combination anti-PD-1/anti-CTLA-4.**
- **Some participants (P3), combination anti-PD1/anti-CTLA-4 was associated with a reduction in HIV DNA and inducible virus.**
- **Combination anti-PD1/anti CTLA-4 was associated with increased HIV-specific immune responses, but only in one participant (P2).**
- **Our results highlight that ICB can activate latent HIV and increase HIV-specific immune responses, but with variation between individuals.**

**Fig 2. Virological assessments.**
A) US-HIV RNA; B) US HIV RNA fold change from baseline to 24 h post infusion; C) total HIV DNA; D) HIV DNA and MS-RNA by TILDA for P3 at baseline and end of study.

**Fig 3. Immunological responses.**
A) flow cytometry plots showing cytokine response to Gag stimulation in effector memory CD8+ T cells for P2 only, B) Proportion of responding cells to ICS