Microbial translocation does not drive immune activation in Ugandan children with HIV

Fitzgerald F.1,1, Lhomme E.2, Harris K.3, Kenny J.1, Doyle R.4, Kitto C.5, Walker A.S.6, Thiebaut R.2, Klei N.1 and the CHAPAS3 Trial Team


Background

Immune activation (IA), potentially driven by microbial translocation (MT), is linked to increased morbidity despite ART in HIV. We investigated MT as a driver of IA in HIV-infected African children.

Methods

ART-naive and ART-experienced children were recruited to a Ugandan site of the CHAPAS3 Trial (ISRCTN96970975), with HIV-uninfected age-matched controls from the same communities. 19 markers (cellular and soluble) of IA, cellular proliferation, inflammation, and disordered thrombogenesis were measured at weeks 0, 48, and 96, alongside viral load at week 96 and CD4 cell count/percentage. Intestinal fatty acid binding protein (I-FABP) was used to quantify gut damage and MT was assessed using a panel of specific bacterial polymerase chain reactions (PCRs), broad-range 16S rDNA PCR and next generation sequencing (NGS;Illumina method), with bespoke method adaptations to enable sequencing of very low bacterial DNA levels. MT and I-FABP were assessed at weeks 0, 12 and 72. Cluster analysis of IA and MT markers was performed in R.

Table 1 Biomarkers of immune activation, cellular proliferation, inflammation and vascular injury

<table>
<thead>
<tr>
<th>Soluble biomarkers of inflammation and vascular injury</th>
<th>Interleukin (IL)-1β</th>
<th>IL-6</th>
<th>Tumour necrosis factor (TNF)-α</th>
<th>Monocyte chemoattractant protein 1 (MCP-1)</th>
<th>Intercellular adhesion molecule 3 (ICAM-3)</th>
<th>C-reactive protein (CRP)</th>
<th>Intestinal fatty acid binding protein (I-FABP)</th>
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</thead>
<tbody>
<tr>
<td>C-reactive protein (CRP)</td>
<td>Serum amyloid A (SAA)</td>
<td>Tissue factor (TF)</td>
<td>Tumour necrosis factor (TNF)-α</td>
<td>Monocyte chemoattractant protein 1 (MCP-1)</td>
<td>Intercellular adhesion molecule 3 (ICAM-3)</td>
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Results

249 children were included: 120 ART-naive and 120 ART-experienced (median [IQR] age 2.8 [1.7-4.0] & 6.5 [5.9-9.2] years; median baseline CD4% 20 [14-24] & 34 [31-39], respectively) and 107 age-matched HIV-uninfected controls.

- Immune recovery was good. ART-naive: median CD4% change 17 [12-21] and viral load suppression <100 copies/mL at 96 weeks was 76% (ART-naive) and 91% (ART-experienced).
- IA decreased over time on ART. By 96 weeks in ART-naive children, median CD4% HLA-DR+CD38- decreased from 7% to 2%; median [IQR] CRP and TNF-α decreased from 6.4 mg/L (2.9-7.3) to 2.5 (0.8-12.0) and 9.3 pg/mL (6.5-11.9) to 4.4 (2.5-9.5) respectively (p<0.001 for all).
- Specific and broad-range PCR results for bacterial DNA were negative/very low in all groups and over time (Figures 1 & 2).
- At baseline using WGS, very low levels of microbial DNA were found in both HIV-infected groups, including Staphylococcus aureus, Enterobacteriaceae, Veillonellaceae and Clostridiaceae.

Discussion

Although there was some evidence of clustering by immune activation markers at week 96, there was no clear clustering by HIV status or VL suppression.

- Levels of bacterial DNA were low regardless of HIV/ART/IA status or on ART.
- MT markers did not appear to be linked to HIV infection, immune status or IA.
- MT may not be a significant driver of IA in this setting.

Conclusions

1. Immune recovery was good with comitant decrease in IA over time on ART
2. Although there was some evidence of clustering by immune activation markers at week 96, there was no clear clustering by HIV status or VL suppression.
3. Levels of bacterial DNA were low regardless of HIV/ART/IA status or time on ART.
4. MT markers did not appear to be linked to HIV infection, immune status or IA.

MT may not be a significant driver of IA in this setting.