Complex Pattern of Inflammatory Biomarkers after ART Initiation in HIV-Infected African Children

Poster 910

Nigel Klein[1], Chipo Berejena[2], Godfrey Pimundu[3], Pietro Pala[4], Tichaona Vhembo[2], Victor Musiime[3], Phillip Kasirye[5], Hannah Poulsom[1], A. Sarah Walker[6], Andrew J. Prendergast[7] and the ARROW Trial Team

[1] Institute of Child Health, London, UK; [2]University of Zimbabwe, Harare, Zimbabwe; [3]Joint Clinical Research Centre, Kampala, Uganda; [4]UVRI/MRC Uganda Research Unit on AIDS, Entebbe, Uganda; [5]Baylor-Uganda, Paediatric Infectious Diseases Clinic, Mulago Hospital, Kampala, Uganda; [6]MRC Clinical Trials Unit at UCL, London, UK; [7]Queen Mary University of London, London, UK.

Contact details
MRC Clinical Trials Unit at UCL
email: rmjlasw@ucl.ac.uk

www.arrowtrial.org

ABSTRACT (updated)

Background: HIV infection is characterized by immune activation, likely driven by viral replication, microbial translocation, malnutrition and co-infections, particularly in sub-Saharan Africa. Although characterized in HIV-infected adults, changes in inflammatory biomarkers have not been well described in African children starting ART with advanced disease.

Methodology: 316 children/adolescents (median (IQR) age 5 (2,9) years, 67% WHO stage 3/4) initiating ART in the ARROW trial in Uganda/Zimbabwe were enrolled in a substudy measuring inflammatory biomarkers (CRP, TNF-α, IL-6, sCD14) by ELISA on cryopreserved plasma at enrolment and then every 24 weeks to 192 weeks. VL was assayed using Abbott Taqman and Roche Amplicor, with lower detection limit 80 c/ml. Log_{10} absolute cytokine levels after ART initiation were modelled using random effects models with child-level intercepts and slopes, adjusting for site performing the assay.

Results: At ART initiation, geometric mean CD4% was 10.2%, VL 209816c/mL, CRP 4.8 mg/L, TNF-α 24.1 pg/mL,IL-6 6.7pg/mL and sCD14 1.9x10^6 pg/mL. Children with lower pre-ART CD4% had higher pre-ART CRP (p=0.004), IL-6 (p<0.001) and sCD14 (p<0.001), but there was no association with pre-ART TNF-α (p=0.66). In contrast, children with higher pre-ART VL had significantly higher TNF-α (p<0.001), higher IL-6 (p<0.001) and higher sCD14 (p<0.001), but a weaker association with CRP (p=0.04). VL suppression <80 c/ml was achieved by 78% children at 24 weeks; then 76%, 67%, 69%, 66% and 64% at 48, 72, 96, 144 and 196 weeks. IL-6 dropped by 36% [95% CI 27-44%] over the first 24 weeks on ART (p<0.0001), but did not change significantly thereafter (p=0.99). CRP dropped by 45% [32-658] over the first 48 weeks on ART (p<0.001), again with no evidence of change thereafter (p=0.64).TNF-α dropped by 75% [71-78%] over the first 48 weeks on ART, but then rose again by 48% [41-56%] to week 72, subsequently declining more modestly by 38% [34-42%] every 24 weeks (all p<0.001). This effect was seen independently in each site conducting assays, and in all pre-ART CD4% groups. sCD14 declined much more slowly throughout follow-up, dropping by only 1% [0-2%] every 24 weeks (p=0.004).

Conclusions: Children starting ART with advanced disease have elevated inflammatory biomarkers, which are related to pre-ART CD4 levels (except for TNF-α). All biomarkers dropped after starting ART, although declines in sCD14 were very slow. IL-6 and CRP tended to drop rapidly and plateau after 24-48 weeks; by contrast, TNF-α showed more variability, perhaps in response to changes in VL. Better understanding changes in inflammatory biomarkers on ART may help to define subgroups of children with residual immune activation, who would benefit from adjunctive treatment strategies.

BACKGROUND

- HIV infection is characterized by immune activation, likely driven by viral replication, microbial translocation, malnutrition and co-infections, particularly in sub-Saharan Africa
- Although characterized in HIV-infected adults, changes in inflammatory biomarkers have not been well described in African children starting ART with advanced disease

METHODOLOGY

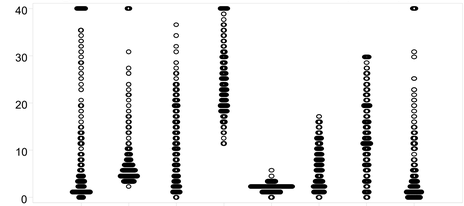
- ARROW (ISCRTN24791884) was a randomised trial of monitoring and first-line treatment strategies among 1206 HIV-infected children in Uganda and Zimbabwe
- **316 Ugandan/Zimbabwean children/adolescents initiating ART** in ARROW were enrolled in a substudy measuring inflammatory biomarkers (CRP, TNF-α, IL-6, sCD14) by ELISA on cryopreserved plasma at enrolment and then every 24 weeks to 192 weeks
  - VL was assayed using Abbott RealTime and Roche Amplicor 1.5, with lower detection limit 80 c/ml (many samples diluted due to low volumes)
  - Log_{10} absolute cytokine levels after ART initiation were modelled using random effects models with child-level intercepts and slopes. Results were similar with (shown) and without (not shown) adjustment for assay site and pre-ART CD4%

RESULTS – ART INITIATION

- Median (IQR) age was 5 (2,9) years; 67% were WHO stage 3/4. See Figure 1 for baseline biomarker levels
- At ART initiation, children with lower pre-ART CD4% had higher pre-ART CRP (p=0.004), IL-6 (p<0.001) and sCD14 (p<0.001), but there was no association with pre-ART TNF-α (p=0.66)
- In contrast, children with higher pre-ART VL had significantly higher TNF-α, higher IL-6 and higher sCD14 (all p<0.001), with only a weak association with CRP (p=0.04)

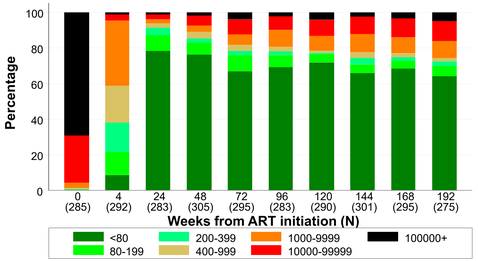
RESULTS

Figure 1: Biomarkers at ART initiation (n=309)



	CRP (mg/L)	IL6 (pg/mL)	IL7 (pg/mL)	TNFα (pg/mL)	sCD14/10^6 (pg/mL)	age (yrs)	CD4%	VL/10^6 (c/ml)
Median	4.0	5.8	9.8	23.9	2.1	5.8	13	228825
Geometric mean	4.8	6.7	7.3	24.1	1.9	4.6	10.2	209816

Figure 2: Virologic suppression after ART initiation



- IL-6 dropped by 36% [95% CI 27-44%] over the first 24 weeks on ART (p<0.0001), but did not change significantly thereafter (p=0.99) (Figure 3)
- CRP dropped by 45% [32-58%] over the first 48 weeks on ART (p<0.001), again with no evidence of change thereafter (p=0.64) (Figure 4)
- sCD14 declined much more slowly throughout follow-up, dropping by only 1% [0-2%] every 24 weeks (p=0.004) (Figure 6), similarly to adults [1]

Figure 3: IL6 after ART initiation (pg/mL)

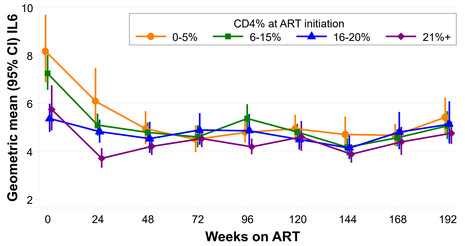
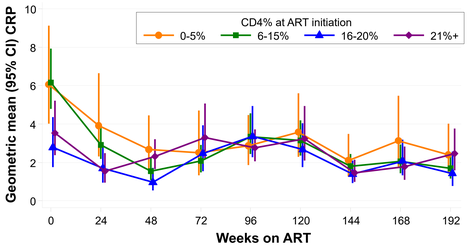


Figure 4: CRP after ART initiation (mg/L)



- TNF-αdropped by 75% [95% CI 71-78%] over the first 48 weeks on ART, but then rose again by 48% [41-56%] to week 72, subsequently declining more modestly by 38% [34-42%] every 24 weeks (all p<0.001) (Figure 5)
- This effect was seen independently in each site conducting assays, and in all pre-ART CD4% groups
  - Possibly related to VL changes between 48 & 72 weeks

References [1] Méndez-Lagares G, et al. *J Infect Dis* 2013;207(8):1221-5

Figure 5: TNFα after ART initiation (pg/mL)

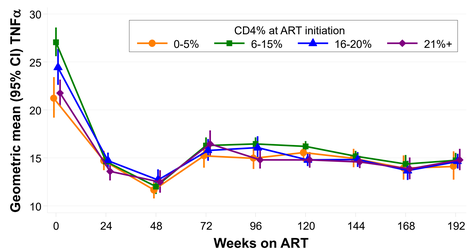
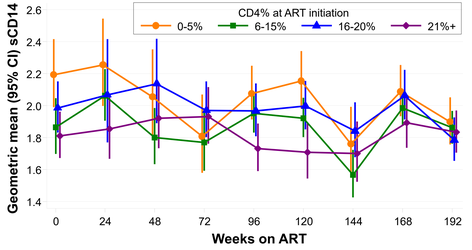


Figure 6: sCD14 after ART initiation (x10^6 pg/mL)



CONCLUSIONS

- **Elevated inflammatory biomarkers in children starting ART with advanced disease normalise quickly on ART, with the exception of sCD14**
  - **more variability in TNF-α over time may reflect VL response**
- **Better understanding of changes in inflammatory biomarkers on ART may help to define subgroups of children with residual immune activation, who would benefit from adjunctive treatment strategies.**

We thank all the patients and staff from all the centres participating in the ARROW trial. Joint Clinical Research Centre, Kampala, Uganda: P Mugyenyi, V Musiime, R Keishanyu, VD Afayo, J Bwomezi, J Byaruhanga, P Erimu, C Karungi, H Kizito, WS Namala, J Namusanje, R Nandugwa, TK Najjuko, E Natukunda, M Ndigendawani, SO Nsiyona, R Kibenge, B Bainomuhwezi, D Sseremba, J Tezikyabbiri, CS Tumusiime, A Balaba, A Mugumya, F Nghania, D Mwebesa, M Mutumba, E Bagurukira, F Odongo, S Mubokyi, M Ssenyonga, M Kasango, E Lutalo, P Oronon; **MRC/UVRI Uganda Research Unit on AIDS, Entebbe, Uganda**: P Munderi, P Nahirya-Ntege, R Katuramu, J Lutaakome, F Nankya, G Nabulime, I Sekamatte, J Kyarimpa, A Ruberantwari, R Sebukyu, G Tushabe, D Wangi, M Musinguzi, M Aber, L Matama, D Nakitto-Kesi; **Baylor-Uganda, Paediatric Infectious Disease Centre, Mulago Hospital, Uganda**: A Kekitiinwa, P Musoke, S Bakeera-Kitaka, R Namuddu, P Kasirye, A Babirye, J Asello, S Nakalanzi, NC Ssemambo, J Nakafeero, J Tikabibamu, G Musoba, J Ssanyu, M Kisekka; **University of Zimbabwe, Harare, Zimbabwe**: KJ Nathoo, MF Bwakura-Dangarembizi, F Mapinge, E Chidziva, T Mhute, T Vhembo, R Mandidewa, M Chipiti, R Dzapasi, C Katanda D Nyoni, GC Tinago, J Bhiri, S Mudzingwa, D Muchabaiwa, M Phiri, V Masore, CC Marozva, SJ Maturure, S Tsikirayi, L Munetsi, KM Rashirai, J Steamer, R Nhema, W Bikwa, B Tambawoga, E Mufuka; **Medical Research Council Clinical Trials Unit, London, UK**:DM Gibb, MJ Thomason, AS Walker, AD Cook, B Naidoo-James, MJ Spyer, C Male, AJ Glabay, LK Kendall, J Crawley, AJ Prendergast. **Independent ARROW Trial Monitors:** I Machingura, S Ssenyonjo. **Trial Steering Committee:**I Weller (Chair), E Luyirika, H Lyall, E Malianga, C Mwansambo, M Nyathi, F Miiro, DM Gibb, A Kekitiinwa, P Mugyenyi, P Munderi, KJ Nathoo, AS Walker; Observers S Kinn, M McNeil, M Roberts, W Snowden. **Data and Safety Monitoring Committee**: A Breckenridge (Chair), C Hill, J Matenga, A Pozniak, J Tumwine. **Endpoint Review Committee**: G Tudor-Williams (Chair), H Barigye, HA Mujuru, G Ndeezi; Observers: S Bakeera-Kitaka, MF Bwakura-Dangarembizi, J Crawley, V Musiime, P Nahirya-Ntege, A Prendergast, M Spyer.

Funding: ARROW was funded by the **UK Medical Research Council** and the **UK Department for International Development** (DFID). **Viiv Healthcare/GlaxoSmithKline** donated first-line drugs for ARROW and provided funding for VL assays.