SUPPLEMENTARY METHODS

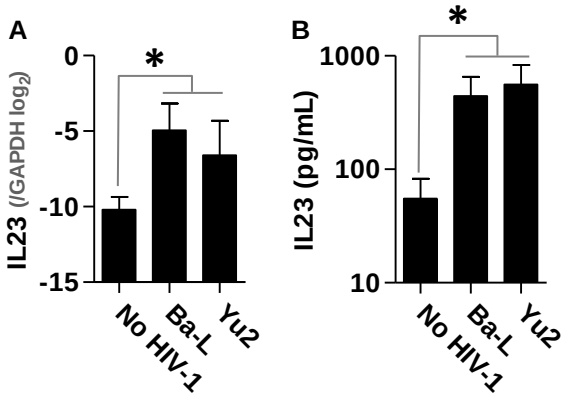
MONOCYTE DERIVED MACROPHAGES

Peripheral blood mononuclear cells (PBMC), consisting of monocytes and lymphocytes, were obtained by density gradient centrifugation of heparinised blood with Lymphoprep (Axis-Shield) according to the manufacturer's instructions. After repeated washing with phosphate buffered saline (PBS) PBMC were resuspended at $1x10^7$ cells/ml in RPMI 1640 with L-glutamine (GIBCO, Invitrogen) containing 5% heat inactivated pooled human AB serum (ABS) (Sigma Aldrich), and seeded on to 6 well tissue culture plates (TPP) at a density of $2x10^6/cm^2$. After one hour at 37°C, non-adherent cells (lymphocytes) were removed by sequential washes with PBS. Adherent cells (predominantly monocytes) were incubated in RPMI 1640 containing 10% heat inactivated autologous serum (AS) supplemented with 20ng/ml M-CSF (R&D Systems). Any remaining non-adherent cells were removed by further washes with PBS and the maturation medium replaced (without any additional M-CSF) on day 3-4. After 6 days of culture, 10% AS was replaced with 5% ABS. This protocol typically yields $1x10^5$ MDM/cm^2.

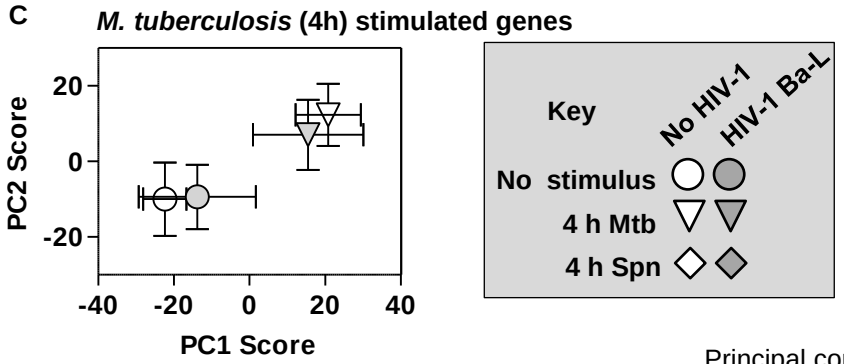
INDUCED SPUTUM AND BRONCHOALVEOLAR LAVAGE PROCESSING

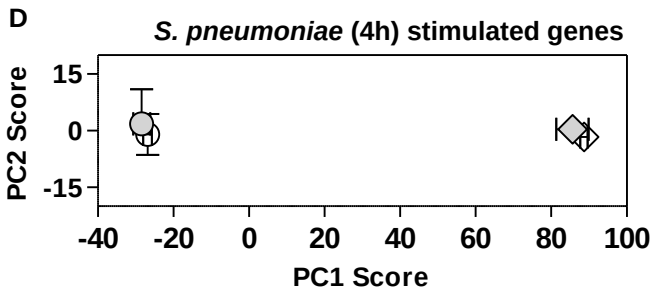
Sputum and bronchoalveolar lavage (BAL) samples were processed within two hours of collection. Firstly, to achieve mucolysis of induced sputum an equivalent volume of 0.1% dithiothreitol solution (Sigma, UK) in PBS was added and the sample rolled for 20 minutes at room temperature. Both BAL and induced sputum samples were then passed through a 100µm filter (Cell Trics™, Partec GmBH) to remove large particulate debris before centrifugation at 430 x g for 10 minutes. The resultant supernatant was removed and stored at -20°C. Samples were sterilised by filtration through a 0.2µm membrane (Millipore) before cytokine analysis.

Supplementary Figure 1

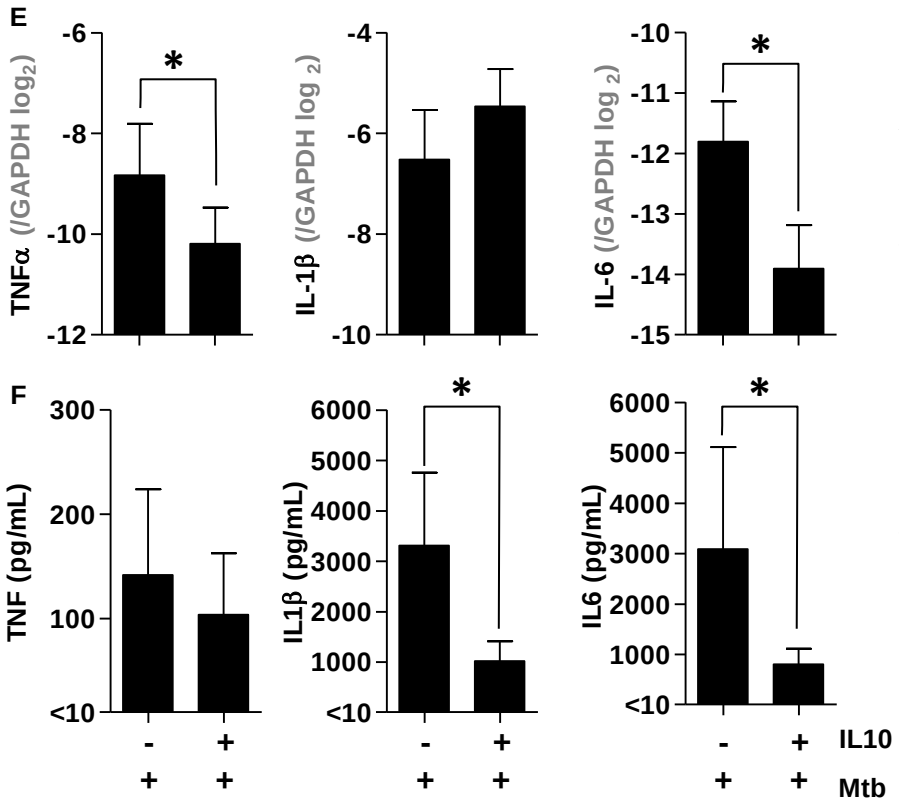


Enhanced inflammatory IL23 responses to Mtb in HIV-1 (Ba-L or Yu2 strains) infected cells were confirmed by qPCR **(A)** and protein ELISA in cell culture supernatants **(B)**. Bars represent mean ± SEM for at least three separate experiments (*denotes p<0.05, t-test).





Principal component (PC) analysis of the major transcriptional responses (represented by PC1 and PC2) to four hour stimulation with Mtb **(C)** or *S. pneumoniae* **(D)** showed comparable changes in HIV-1 infected and uninfected MDM.



Addition of recombinant IL10 (10 ng/mL) to MDM significantly attenuated TNFα and IL6 transcriptional responses quantified by qPCR after 24 hour stimulation with Mtb **(E)**, and IL1β and IL6 protein responses quantified by ELISA after 72 hours stimulation with Mtb **(F)**. Bars represent mean ± SEM for at least three separate experiments (*denotes p<0.05, t-test).

SUPPLEMENTARY TABLE 1

TaqMan Gene Expression Inventoried Assays

Gene	Applied Biosystems Assay ID
CCL20	Hs01011368_m1
IL23	Hs00372324_m1
IL1β	Hs01555410_m1
IL10	Hs99999035_m1
IL6	Hs00985639_m1
TNFα	Hs00174128_m1
HPRT1	Hs99999909_m1
HIV-1 LTR	Pa03453409_s1