IL-4 enhances IL-10 production in Th1 cells: implications for Th1 and Th2 regulation

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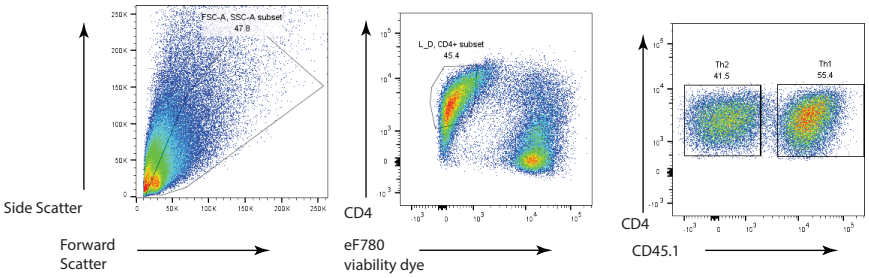
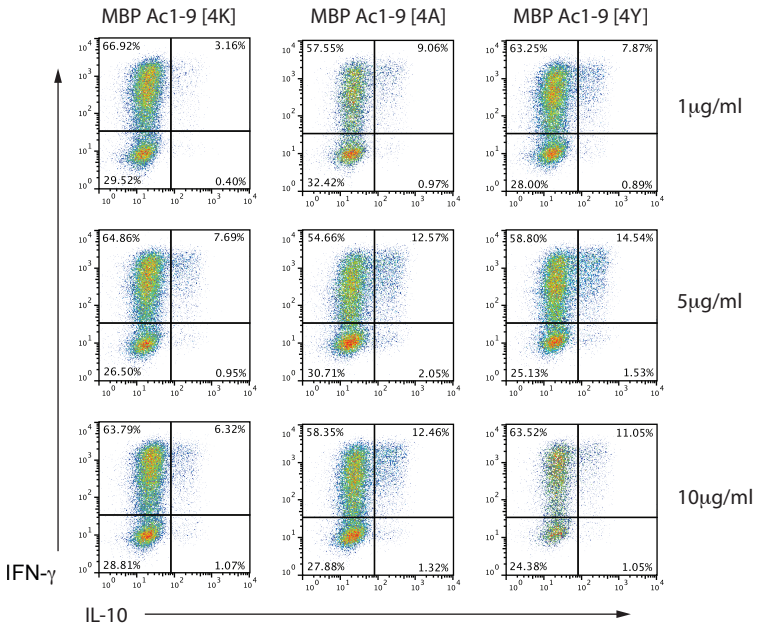


Supplementary Figure S1: CD4+ T cell gating strategy

An example is shown of the FACS gating strategy used. Lymphocytes were gated in a FSC/SSC plot as indicated and live $CD4^+$ cells within this population were gated in a CD4-Alexa700 / ef780 viability dye plot as indicated. If in a co-culture of Th1 and Th2 cells, cells were gated on $CD45.1^+$ cells to identify the Th1 cells. These cells were then further analysed for intracellular cytokines or transcription factors.



Supplementary Figure S2: The affinity and dose of peptide determine the differentiation of IL-10 secreting CD4$^{\pm}$ cells

Splenic Tg4 CD4^{+} T cells were cultured in the presence of irradiated B10.PL splenocytes as APCs and MBP Ac1-9 peptide affinity variants (MBP Ac1-9[4A], MBP Ac1-9[4K], MBP Ac1-9[4Y]) at a number of different concentrations (1µg, 5µg, 10µg). Intracellular cytokine staining was carried out on day 7 of the second stimulation, following PMA and ionomycin stimulation. FACS plots from a single experiment are gated on live CD4+ cells and show IFN-γ, IL-4 and IL-10 expression.