



**Figure 5—figure supplement 3:** TNF- $\alpha$  skews polarization of monocytes to macrophages with lower CD80 expression. PBMCs were infected with Mtb H37Rv at MOI of 0.1 and encapsulated in alginate-collagen microspheres after overnight incubation. Microspheres were then incubated in complete RPMI (with L-Glutamine and 10% human serum) with TNF- $\alpha$  7.5ng/ml. Uninfected PBMCs were encapsulated and treated similarly as a comparator for TNF- $\alpha$  stimulation. At day 7, the microspheres were decapsulated in 0.5mM EDTA solution at pH of 7.2. Double staining with CD14 and CD11b defined macrophages, which were classified by CD80 and CD163 expression. (A) Histogram showing expression of CD163 and CD80 where there was significant decrease in CD80 expression as shown in Figure (B). TNF suppressed the relative geometric mean of CD80, but did not affect CD163 expression. This experiment was performed in 4 separate donors.