

Figure S5. Inhibition of HDAC activity during monocyte differentiation modifies transcript levels of several cytokines/ chemokines in Mo1 upon Mtb infection. A. Outline of the experimental setup used in S5B-D. B. Monocytes derived from 4 different donors were differentiated towards M\(\phi \) while being exposed to TMP195 (300 nM), TMP269 (300 nM), TSA (30 nM) or DMSO at equal v/v for 6 days. Differentiated Mφ1 were subsequently infected with Mtb for 1h at MOI 10 and incubated for 48 hours with different amounts of IFN-γ. Dots depict the median bacterial survival of 4 donors expressed as a percentage of the DMSO control while whiskers represent 95% confidence intervals. Statistically significant differences were tested using a RM one-way ANOVA with repeated measures and Dunnett's multiple test correction. C. Cell viability measurement of Mtbinfected M ϕ 1 (experimental setup as in A). Dots represent the mean of 2 viability assay replicates of a single donor expressed as a percentage of the DMSO control. Bars indicate median values of all 4 donors and whiskers represent 95% confidence intervals. Statistically significant differences were tested using a RM one-way ANOVA. D. Transcript levels of HDAC1 and HDAC5 were determined by qPCR in duplicate in Mtb-infected Mφ1 from 5 different donors exposed to TMP195 (300 nM), TMP269 (300 nM), TSA (30 nM) or DMSO at equal v/v during differentiation. Data was normalized to GAPDH ((2-ΔCT(HDAC))/(2-ΔCT(GAPDH))) and mean expression levels of duplicate samples were calculated for each donor. Box-andwhisker plots (min. to max.) show gene expression levels of the 5 donors where each dot represents a single donor. Significant differences between treatments were determined using a RM one-way ANOVA with Dunnett's multiple test correction. E. Transcript levels of HDAC1 and HDAC5 were determined by qPCR in duplicate in Mtb-infected Mφ1 from 5 different donors in the presence or absence of IFN-γ (1000 pg/ml). M 1 had been exposed to TMP195 (300 nM), TMP269 (300 nM), TSA (30 nM) or DMSO at equal v/v during differentiation. Data was normalized to GAPDH and mean expression levels of duplicate samples were calculated for each donor. Dot plots display log₂ FC expression levels to their respective baseline controls (Mφ1 in the absence of IFN-γ) calculated using the 2-ΔΔCT formula. Significant differences between presence and absence of IFN-γ were determined using a paired sample t-test (* = p<0.05, ** = p<0.01).