Figure S1. Foxp3 acetylation and increased expression does not depend on homodimerization

(A) Flag-Foxp3 or Flag-Foxp3ΔE250 was co-transfected with p300 in HEK 293 cells. Using equal amounts of input protein, Foxp3 acetylation was analyzed using antibodies specific for acetyl-lysine (Ac-Lys), Flag or HA. (B) Cells were co-transfected with Flag-Foxp3 or Flag-Foxp3 ΔE250 and/or p300 and treated with 100 nm TSA and 2.5 mM NAM for 16 hours. Protein lysates were quantified and subsequently western blots were probed utilizing an antibody recognizing Flag, HA, or tubulin as loading control. All western blots are representative of at least 3 independent experiments.
Figure S2. Increased Foxp3^+ cell numbers after HDACi treatment is not due to preferential cell death
(A) Human CD4^+ cells from six different donors were cultured in RPMI medium in the presence of IL2, anti CD3 and anti CD28 with or without NAM as described in figure 5 and 6. At day 7 cells were stained for FOXP3 to separate Foxp3^- from Foxp3^+ cells and annexin V and 7-AAD to determine the percentage of apoptosis. The ratio of apoptotic cells in FOXP3^+ vs FOXP3^- is depicted. (B) Analysis of Foxp3 positivity in CD4^+ compartment of human PBMCs at day 0 as the cells go into culture.