



Figure 7- figure supplement 2. Influence of ThPOK on the differentiation of cytotoxic CD8 T cells. Naive CD8 T cells were submitted to polyclonal stimulation with PHA in the presence of irradiated allogeneic feeder cells, IL-2, IL-15 and Th1 polarizing cytokines for 7 to 14 days. Perforin, granzyme B (GZMB) and ThPOK expression was measured by flow cytometry and by qPCR. **a)** Differentiated CD8 T cells expressed perforin and ThPOK. Gates were placed using an unstained control and numbers indicate the % of cells in each quadrant. Similar results were obtained in three independent experiments. **b)** Suppression of ThPOK expression in differentiated CD8 T cells was performed using specific shRNA as done in Figure 7. ThPOK knockdown, verified at mRNA and protein levels, did not significantly influence the expression of perforin but significantly increased the expression of granzyme B as compared to non-silencing (N-S) shRNA. Data are mean \pm SD log₂ fold change of 5 replicates from 3 independent experiments (upper panels) and one histogram with median fluorescence intensity (MFI) of 2 independent experiments. *:p<0.05. These results support the notion that ThPOK expression could limit the cytotoxic function of CD8 T cells, as observed in CD4 T cells.