## Piexoto et al., http://www.jem.org/cgi/content/full/jem.20062349/DC1

## SUPPLEMENTAL TEXT

## Cell screening

We initially screened CD8 T cells recovered at different points of the immune reaction for the expression of 14 effector genes, and we found that the  $T_{H2}$  cytokine genes (*Il4, Il5,* and *Il13*), *Lta,* and *Il15* were never expressed, and that *Il10* expression was extremely rare. At some time points, we could find a single cell scoring positive for this cytokine, but, more frequently, all cells were negative. *Il2* mRNA expression was relatively more abundant because two to three cells per time point were usually scored. The expression frequency of these rare genes is too low to allow accurate evaluation at a single-cell level and is not described further.

## Cell coexpression

We enumerated the secondary memory cells coexpressing *Prf1 and Gzmb* (and thus potentially able to use the efficient perforin killer pathway), as well as those coexpressing *Fasl*, which thus have the potential to use both perforin and Fas-L killer pathways simultaneously. We found 15% of the former and an additional 13% of the latter cell types in SM-CD8s. Thus, SM-CD8s had  $\sim$ 28% of cells potentially able to mediate effective killing. In contrast, in PM-CD8s only 3/45 cells coexpressed *Prf1* and *Gzmb*, and 1/45 PM-CD8 cells coexpressed *Prf1 Gzmb* and *Fasl* (Fisher's exact test, P < 0.01).