

Stability of T_H1/T_c1 commitment to an effector lineage

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Polarized T cells that have undergone multiple cell divisions in vitro achieve a terminally differentiated state in which the T cell can no longer alter its cytokine profile, even when stimulated in opposite polarizing conditions. However, the physiological relevance of these findings for T cell lineage commitment in vivo remains unclear. To address this question in vivo, an LCMV (lymphocytic choriomeningitis virus) infection model highly polarizes LCMV glycoprotein-specific CD4 (Smarta Tg⁺) and CD8 (P14 Tg⁺) T cells to T_H1 and T_c1 effector cells, respectively. Smarta Tg⁺ CD4 and P14 Tg⁺ CD8 T cells are CFSE labeled to track cell divisions, adoptively transferred into C57BL/6 recipient hosts, and infected with the Armstrong strain of LCMV to drive polarization to an effector T_H1/T_c1 phenotype. At the peak of the immune response (Day 7-8 post-infection), the transferred T cells are harvested from the LCMV-infected mice and stimulated with APC + peptide in T_H0 (IL-2 only)- or T_H2 (IL-2 + IL-4 + α -IL-12 + α -IFN- γ)-polarizing conditions for one week. Intracellular cytokine staining, ELISA, and RT-PCR are used to assess the production of IFN- γ and IL-4.

Preliminary data indicate that T_H1/T_c1 cells stimulated in vivo display more plasticity than T cells polarized in vitro. Polarized T_H1/T_c1 cells that have undergone multiple cell divisions retained the ability to alter their cytokine profile when stimulated in T_H2/T_c2-polarizing conditions in vitro. When analyzed directly ex vivo or after one week in non-polarizing (T_H0/T_c0) conditions, LCMV-specific CD4 and CD8 T cells produced IFN- γ but did not produce IL-4. However, when stimulated in T_H2/T_c2-polarizing conditions, LCMV-specific CD4 and CD8 T cells repressed their production of IFN- γ while IL-4 message, as determined by RT-PCR, increased.

The in vivo findings raise the possibility that commitment of antigen-specific CD4 and CD8 T cells to an effector lineage may be less stringent than what has been demonstrated in vitro. Future studies will address the stability of memory T_H1/T_c1 cells and secondary effectors to commit to an effector lineage. Additionally, future questions will attempt to understand why antigen-specific T_H1/T_c1 cells polarized in vivo display more plasticity than T_H1/T_c1 cells stimulated in vitro. Cytokine signals provided in vivo might allow for decreased resistance to terminal differentiation into an effector phenotype. These signals may affect the expression of transcriptional activators such as GATA-3 or T-bet or contribute to the methylation/acetylation status of the *ifng* and *il4* loci.