THYMOCYTE DEVELOPMENT

Making a commitment

Common precursors in the thymus can develop into either conventional αβ T cells or intestinal γδ T cells. The developmental stage at which lineage commitment occurs and the role of T-cell receptor (TCR) signalling in this process have been controversial issues. Harald von Boehmer and colleagues have now shown that commitment is instructed by TCR signals.

Double-negative (DN) thymocytes undergo rearrangement of the genes encoding the β, δ and γ chains of the TCR to generate a pre-TCR (containing the β-chain and pTα) or γδ TCR on the cell surface. Pre-TCR-expressing cells, but not most γδ TCR$^+$ thymocytes, then become CD4$^+$CD8$^+$ double-positive (DP) cells in a process known as β-selection, which marks an irreversible commitment to the αβ T-cell lineage. However, some cells expressing a γδ TCR can also progress to the DP stage, whereas other cells that prematurely express an αβ TCR do not become DP. The αβ and γδ lineages are therefore defined on the basis of whether or not cells progress to the DP stage, with the precise role of the different TCRs being unclear.

Using bulk cultures of γδ TCR$^+$ DN thymocytes co-cultured with OP9–DL1 stromal cells (which form an artificial thymic environment), von Boehmer and colleagues showed that 4% of these cells had developed a DP phenotype after 7 days. Previous reports have indicated that strong TCR signals favour γδ T-cell development; in support of this, when the culture wells were coated with a γδ-TCR-specific antibody, DP cells did not develop in the cultures and did not appear after transfer of cells to uncoated wells, which indicates stable γδ-lineage commitment.

Therefore, immature γδ thymocytes can give rise to both αβ and γδ lineages, with the relative lineage proportions depending on the strength of the TCR signal. However, can the progeny of a single cell be instructed by TCR signalling to adopt either fate or is a cell pre-committed to a particular lineage before TCR expression, with TCR signalling acting to select a subpopulation of pre-committed cells? Clones derived from single immature γδ TCR$^+$ cells were split in parallel into antibody-coated or uncoated wells with OP9–DL1 cells; 82% of the uncoated wells contained γδ TCR$^+$ DN cells and 18% contained DP αβ-lineage cells, whereas all of the coated wells contained only DN γδ-lineage cells. Therefore, the 18% of γδ TCR$^+$ clones that would otherwise become DP αβ-lineage cells could be diverted by antibody-mediated TCR signalling to the γδ lineage, which indicates that these cells are not committed to either lineage before TCR expression. Furthermore, analysis of clones derived from single thymocytes that had progressed beyond the β-selection checkpoint and were destined to become αβ-lineage cells showed that they could be inhibited from becoming DP cells by culture in OP9–DL1-containing wells coated with CD3-specific antibody, thereby providing a strong TCR signal. These cells took on a stable DN γδ-lineage phenotype (with expression of a γδ or αβ TCR depending, respectively, on whether antibody was added to the culture before or after αβ TCR expression had been initiated).

The authors therefore conclude that, for the progeny of a single DN thymocyte, commitment to αβ and γδ lineages occurs after TCR expression. A strong TCR signal can induce conversion from the αβ to γδ lineage, with the resulting DN cells expressing either a γδ or αβ TCR depending on when the TCR signal is received.

Kirsty Minton

ORIGINAL RESEARCH PAPER Kreslavsky, T. Garbe, A. I., Krueger, A. & von Boehmer, H. T cell receptor-instructed αβ versus γδ lineage commitment revealed by single-cell analysis. *J. Exp. Med.* 28 April 2008 (doi: 10.1084/jem.20072425)