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Thymopoiesis in 4 dimensions

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Abstract

Developing T cells undergo long-range migrations in the thymus that are tightly linked to their developmental program. In this review, we discuss the anatomical positioning of developing T cells in the thymus, review what we know about how cells move in the thymus, and point out unresolved questions. We also discuss new imaging technologies and interdisciplinary approaches and discuss how the promise they offer to addressing these questions.

Keywords: Thymocyte; Cell migration; Thymic architecture; Development

1. Introduction

Thymopoiesis involves the tightly regulated movement of cells over considerable distances to achieve the final production of mature T cells. In this review we focus on what is known, and not known, of the spatial organization within the thymus, how thymic compartmentalization is maintained, and how cell movement proceeds across the thymic landscape and in concert with development. We also discuss new imaging technologies and interdisciplinary methodologies and the promise they offer in addressing many of the spatial–temporal aspects of thymocyte development.

2. Thymus ontogeny and compartmentalization

Thymic organogenesis begins on E10.5 as the result of epithelial–mesenchymal interactions between the third pharyngeal pouch endoderm and neural crest-derived mesynchyme [1]. Seeding of the thymus anlage by hematopoietic precursors begins on E11.0 [2,3] followed by further phenotypic maturation of thymic epithelium mediated by Wnt-induced expression of the FoxN1 gene [4–6]. Commitment of hematopoietic precursors to the T lineage is established by E14.5 and marks the transition from thymocyte-independent to thymocyte-dependent epithelial development. Beyond this point of development, patterning and differentiation of thymic epithelium occurs in parallel with and is fully dependent upon further thymocyte development [7,8].

By 3 weeks of age the adult thymus is compartmentalized with a DC-rich boundary separating an inner region of medullary tissue from the outer cortex (Fig. 1). The two major regions contain distinct types of stromal elements and there is a considerable degree of epithelial heterogeneity within the compartments, particularly within the medulla [9,10]. Little is known as to how cortico-medullary boundaries are maintained, but recently it has been shown that Eph-A receptors and their ephrin ligands are expressed in thymus. Given their differential expression in thymic stromal subsets [11,12], and their known role in maintaining compartmentalization in other developmental contexts [13,14] it is tempting to speculate that they play a role in maintaining tissue boundaries in the thymus.

3. Thymocyte trafficking

3.1. From bone marrow to thymus

A developing thymocte travels thousands of microns through the thymus before becoming a mature functional T cell. Before entering the thymus, progenitors are released

Abbreviations: DN, double-negative ($CD4^-CD8^-$); DP, double-positive ($CD4^+CD8^+$); SP, single-positive ($CD4^+CD8^-$ or $CD4^-CD8^+$); CMJ, (cortical/medullary junction); SCZ, subcapsulary zone

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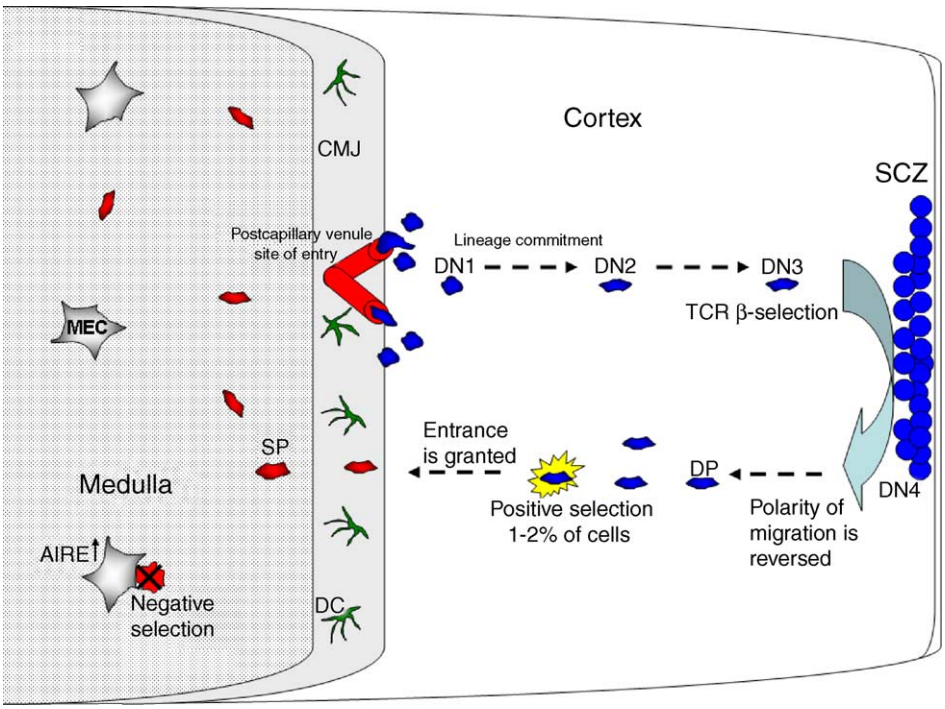

Fig. 1. Intrathymic migration of developing thymocytes. T lineage progenitors enter the thymus through post-capillary venules of the corticomedullary junction (CMJ) and then migrate outward to the subcapsulary zone (SCZ) where they undergo TCR beta chain rearrangement and selection. At this time polarity of migration is reversed and $CD4^+CD8^+$ (DP) thymocytes move back out into the cortex for alpha chain rearrangement and the testing of nascent TCR through positive selection. Post-selected thymocytes are granted access to the medulla where they are screened for self-reactivity through negative selection.

from the bone marrow, circulate through the bloodstream, and ultimately seed the thymus, entering via post-capillary venules within or near the cortico-medullary junction (CMJ) [15] (Fig. 1). The homing process is not continuous, but involves the intermittent release of thymic progenitors from the bone marrow, followed approximately 1 week later by a period of receptivity to thymic seeding [16]. This cyclic mobilization and import of progenitors into the thymus leads to the generation of discrete, overlapping waves of T cell production with a periodicity of approximately 3–5 weeks [17].

The phenotypic identity of the cell that is induced to leave the bone marrow is not known, but a recent report provides evidence that this cell is contained within a primitive population (Lin^- $Sca\text{-}1^+$ $c\text{-}kit^{hi}$ $Thy1.1^-$) and can be distinguished by L-selectin expression [18]. Cells with this phenotypic profile were shown to possess developmental potential that was biased to the T lineage and were phenotypically similar to the early thymic progenitor (ETP) found in thymus. Furthermore, this subset reconstituted the T lineage with kinetics similar to the ETP upon transfer into sublethally irradiated mice. These findings led authors to propose that L-$selectin^+$ cells within the $Lin^-Sca\text{-}1^+$ $c\text{-}kit^{hi}$ $Thy1.1^-$ population were the bone marrow precursors to the ETP found in thymus. Further studies, such as single cell assays, are needed to determine whether this bone marrow subset represents a pure population of thymic progenitors or whether it is a heterogeneous population, which contains the thymic progenitor. In addition, there may be multiple paths that lead from the bone marrow to the thymus and to commitment to the T lineage [19–23].

Upon reaching the thymus through the bloodstream, early progenitors must extravasate through the vascular endothelium of post-capillary venules to enter the organ and then seed the tissue. The factors that confer competency for these events are poorly understood, but a recent report has shown that the small GTPase RAP1 mediates the extravasation of mature T cells across vascular endothelium by redistributing CD44 and CXCR4 within the membrane of extravasating cells [24]. Since both CD44 and CXCR4 are expressed by early thymic immigrants [25–28], and since CD44 is required for proper thymic homing [29,30] it is possible that RAP1 might also mediate the extravasation events of thymic progenitors.

After entry into the thymus, new immigrants must be competent to undergo interactions with their local microenvironment, which ultimately lead to their proliferation and further differentiation to the T lineage. L-selectin ligands, ICAM-1, VCAM-1, and VAP-1 have recently been implicated in the homing process at the local level of thymic seeding [31]. Why the thymus is periodically receptive to seeding is not clear, but windows of receptivity could arise from a limited number of supporting microniches that empty in a synchronous or semi-synchronous niches way. Alternatively, periodic receptivity may reflect the cyclic production of factors that mediate the homing process.