Comment on Dudakov et al, page 933

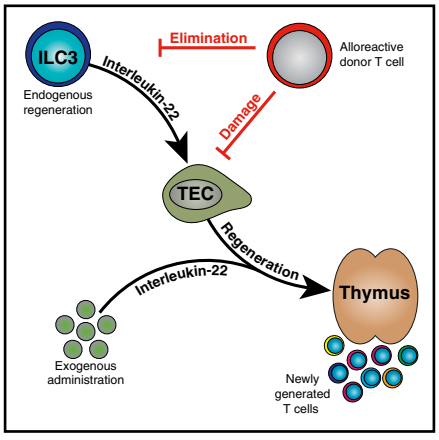
Innately interesting interactions

Krishna V. Komanduri UNIVERSITY OF MIAMI

In the current issue of *Blood*, Dudakov et al present interesting studies of experimental graft-versus-host disease (GVHD) occurring in experimental murine models of allogeneic transplantation, highlighting the importance of dysregulation of the thymus in this setting.[1]

The modern era of allogeneic hematopoietic cell transplantation (alloHCT) began in 1957, several years before Miller and others first proposed the thymus as an important site of leukocyte protection in humans in the early 1960s.[2] Despite a series of studies in the 1960s confirming its significance in lymphopoeisis, the thymus was thought by many to be essentially vestigial beyond childhood until 2 studies in the late 1990s demonstrated definitively that human thymopoiesis persists throughout life.[3,4]

Although maintaining thymopoiesis throughout life to replenish the T-cell repertoire makes logical and evolutionary sense,[5] many students of medicine (and even immunology) continue to be miseducated about this fact. One critical reason thymopoiesis remains misunderstood is that even less is understood regarding how thymic function is dysregulated in disease states, contributing to lymphopenia and autoimmunity; the present study helps to address this.



Overview of interactions between ILC3 and thymic epithelial TECs in the setting of GVHD following alloHCT. Following thymic injury, IL-22 derived from ILC3 or administered exogenously promotes the regeneration of thymopoiesis. See Figure 6D in the article by Dudakov et al that begins on page 933.

Despite persistent ignorance about thymopoiesis, early clinical studies hinted at the importance of the thymus in alloHCT recipients. In 1987, Müller-Hermelink et al[6] studied pathologic specimens of deceased human alloHCT recipients and suggested that multiple processes, including GVHD, contributed to atrophy of the thymus after alloHCT. Much later, armed with quantitative approaches, we directly assessed thymopoiesis in alloHCT recipients and demonstrated that the pace of immune recovery correlated with direct measures of thymopoietic recovery, clearly influencing patterns of T-cell recovery.[7] The current study links thymopoiesis and GVHD while demonstrating the importance of innate lymphoid cells (ILCs) in these processes.

Given their recent appearance on the immunological scene, you will be forgiven if you are not yet familiar with ILC development and classification (reviewed nicely in Artis and Spits[8]). Broadly, ILCs have classical lymphoid morphology and include natural killer cells, which have cytotoxic functions. but no rearranged antigen receptors like T cells.[8] There are also 3 noncytotoxic ILC subsets characterized by their patterns of development and production of effector cytokines when mature. These noncytotoxic ILCs appear to have important roles in immunity to pathogens, as well as tissue homeostasis. The ILC3 group includes a subset of lymphoid tissue inducer cells that have embryological significance in the formation of secondary lymphoid tissues, including the lymph nodes and thymus.[8] ILC3 development is governed by the expression of the aryl hydrocarbon receptor (AHR), whereas their function appears to depend on the expression of the transcription factors RORγt and T-bet.[8] They are further characterized by their production of cytokines including lymphotoxin, interleukin-17 (IL-17), IL-22, and tumor necrosis factor-α, demonstrating their parallels to $CD4^+$ T cells.[8]

Previous studies by this group highlighted the importance of IL-22 produced by ILC3 in intestinal regeneration in the setting of GVHD.[9] That study demonstrated that ILC production of IL-22 was necessary for the proliferation of intestinal stem cells, thereby regulating the maintenance of intestinal architecture in the setting of GVHD. By demonstrating links between ILC and nonimmunological epithelial subsets, this study demonstrated the importance of cellular crosstalk in protection and recovery from the tissue injury that characterizes GVHD.[9]

In the current study, Dudakov and colleagues turned their attention to the thymus. They first demonstrated that, in mice experiencing GVHD after alloHCT, ILC3s were depleted in association with decreased intrathymic levels of IL-22.[1] One consequence of decreased IL-22 levels was decreased numbers of cortical and medullary thymic epithelial cells (TECs), a cellular population that expresses the IL-22 receptor and that is critical for the maintenance of lymphoid cells, including $CD4^+CD8^+$ double-positive thymocytes that are the dominant thymic lymphoid subpopulation.[1] When performing transplants into $IL\text{-}22^{(-/-)}$ recipient mice, they found increased severity of GVHD-associated thymic injury, including stromal damage.[1]

Because IL-21 signaling plays a role in alloreactive T-cell–mediated thymic injury, the authors next examined the thymic effects of GVHD in recipients of transplants from IL-21 receptor knockout mice.[1] Not surprisingly, they saw increased numbers of cortical and medullary TECs; critically, this restoration of thymic architecture was not observed in transplants from IL-21 receptor knockout donors into $IL\text{-}22^{(-/-)}$ recipients. Finally, the authors administered IL-22 to recipient mice shortly after alloHCT; consistent with their prior observations, IL-22 administration significantly improved thymopoiesis as measured by the direct output of recent thymic emigrants.[1]

This study highlights the role of 2 cytokines with somewhat opposing actions: IL-21 produced by alloreactive donor T cells, a mediator of thymic injury; and ILC3-derived

IL-22, which they confirm has cytoprotective functions for thymic epithelia (see figure). This study also highlights the importance of cellular crosstalk between ILCs, TECs, and developing lymphoid cells in the restoration of function of an organ that is essential for normal T-cell homeostasis.

Although the authors tell a compelling and convincing story through their work, questions do remain. The models used in the Dudakov et al study, as is typical for murine transplantation, used radiation-based conditioning. ILCs are relatively radioresistant, and it will be important to understand how conditioning approaches in the human setting (typically less intensive and cytotoxic chemotherapy based) will influence ILCs, TECs, and the crosstalk observed in these models. Murine transplants are typically performed with young mice, which more closely resemble pediatric (rather than adult) recipients in terms of lymphoid and thymic architecture and immune recovery. In general, these events will be very difficult to confirm in humans, given the practical impossibility with current technologies of assessing rare cellular subpopulations and cytokine levels within secondary lymphoid organs.

Despite these limitations, the studies performed by Dudakov et al are important and suggest a path toward clinical translation. Indeed, it may be easier to assess whether IL-22 improves thymic recovery in humans than to confirm whether the events seen in murine models precisely replicate human alloHCT pathophysiology. Furthermore, other approaches (eg, modulation of AHR pathways) might also prove useful in maintaining or restoring thymopoiesis. Ultimately, it is reassuring to see so much progress in understanding how to restore the function of an organ so important and yet so poorly understood. This work should inspire additional studies and, I hope, meaningful therapies.

Conflict-of-interest disclosure: The author declares no competing financial interests. ■

REFERENCES

1. Dudakov JA, Mertelsmann AM, O'Connor MH, et al. Loss of thymic innate lymphoid cells leads to impaired thymopoiesis in experimental graft-versus-host disease. *Blood*. 2017;130(7):933-942.

2. Miller JFAP. Immunological function of the thymus. *Lancet*. 1961;2(7205):748-749.

3. Douek DC, McFarland RD, Keiser PH, et al. Changes in thymic function with age and during the treatment of HIV infection. *Nature*. 1998;396(6712):690-695.

4. Poulin JF, Viswanathan MN, Harris JM, et al. Direct evidence for thymic function in adult humans. *J Exp Med*. 1999;190(4):479-486.

5. Komanduri KV, McCune JM. Development and reconstitution of T-lymphoid immunity. In: Pantaleo G, Walker BD, eds. Retroviral Immunology: Immune Response and Restoration. Totowa, NJ: Humana Press; 2001:79-107.

6. Müller-Hermelink HK, Sale GE, Borisch B, Storb R. Pathology of the thymus after allogeneic bone marrow transplantation in man. A histologic immunohistochemical study of 36 patients. *Am J Pathol*. 1987;129(2):242-256.

7. Komanduri KV, St John LS, de Lima M, et al. Delayed immune reconstitution after cord blood transplantation is characterized by impaired thymopoiesis and late memory T-cell skewing. *Blood*. 2007;110(13):4543-4551.

8. Artis D, Spits H. The biology of innate lymphoid cells. *Nature*. 2015;517(7534):293-301.

9. Lindemans CA, Calafiore M, Mertelsmann AM, et al. Interleukin-22 promotes intestinal-stem-cell-mediated epithelial regeneration. *Nature*. 2015;528(7583):560-564.

DOI 10.1182/blood-2017-06-791848