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INSIGHTS

Allo-reactive tissue-resident T cells causing damage: An inside job

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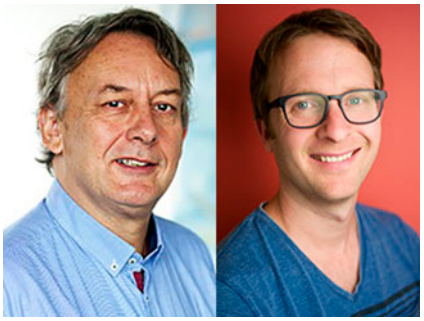
Tissue-resident memory T cells (T_{RM} cells) reside in the epithelium and contribute to the first line defense against invading pathogens. Snyder et al. (2022. *J. Exp. Med.* https://doi.org/10.1084/jem.20212059) now report that clonally expanded, recipient T cells persist as T_{RM} cells in human lung allografts despite intensive immunosuppression. Their persistence may contribute to chronic allograft dysfunction.

Much of our current understanding on the role of tissue-resident memory T cells (T_{RM} cells) in immune protection and immunopathology stems from experimental mouse models in which T_{RM} can unequivocally be identified, followed, and manipulated. For obvious reasons, sampling of human tissue is more problematic but at the same time essential to enhance our understanding of the role of T_{RM} cells in human pathology (Piet et al., 2011; Hombrink et al., 2016; Snyder et al., 2019; Gray and Farber, 2022). In this context, the meticulous analyses Snyder and colleagues performed on three patients after receiving a lung allograft provide important indications on the contribution of T_{RM} cells to acute and chronic graft (dys)function (Snyder et al., 2022). The data presented raise a number of key points: the impact of T_{RM} cells on pathology cannot be determined from blood analyses, T_{RM} cells respond to but cannot be removed by glucocorticoids, and, perhaps most importantly, we do need better and specific tools to alter the behavior of T_{RM} cells in patients.

A considerable fraction of lung transplantation patients suffers from acute cellular rejection (ACR). The authors found, in tissue obtained via transbronchial biopsies at the time of ACR, perivascular T cell infiltrates mainly of recipient origin. Immunophenotyping of T cells obtained from the bronchoalveolar lavage (BAL) showed a predominance of effector memory $CD4^+$ and $CD8^+$ T ($CCR7^-CD45RA^-$) cells. The BAL contained clonally expanded $CD8^+$ T cells at the time of ACR; some of them were demonstrated to be allo-reactive. At the transcriptional and protein level, these clonal populations appeared to be able to execute immediate effector functions as they not only expressed molecules involved in cytotoxicity (*GZMB*, *GZMK*, *PRF1*) but also IFNγ. Furthermore, the upregulation of genes associated with tissue residency (*ITGAE*, *ITGA1*, *PRDM1*, *CXCR6*, *LAG3*) and downregulation of genes involved with tissue egress (*CCR7* and *S1PR1*) suggest that these clonal populations are constituted of true T_{RM} cells. In line with the notion that within the transplant tissue, T cells clonally expand, acquire canonical T_{RM} cell features, and as a consequence take residency. Further, clones that are predominant in the lung were hardly found in the circulating pole. Finally and importantly, it was found that expanded clones persist for up to half a year after systemic glucocorticoid therapy for ACR. After suppression of ACR by methylprednisolone, however, recipient-derived T_{RM} cells reduced the expression of genes involved in effector functions such as granzymes, perforin, and IFNγ. Taken together, the data presented provide compelling evidence that allo-reactive T cells that have been recruited to the lung clonally expand and persist as resident effector T cells. Although the effector functions of these in-part allo-reactive T_{RM} cells can be downregulated by high-dose glucocorticoids, they persisted in spite of treatment. The latter observation is likely to be meaningful in understanding the pathophysiology of long-term allograft lung damage.



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After lung transplantation, recipient T cells gradually acquire T_{RM} cell phenotypes (Snyder et al., 2019). In reminiscence of some Wall Street bankers featured in the film *Inside Job*, when the intruding and expanding recipient T cell clones acquire dominance over the donor-derived lung T_{RM} cells, homeostasis is disturbed and damage may be inflicted to the allograft. As T_{RM} cells

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largely remain at distant epithelial sites, their (organ-)specific properties in (patho) physiology cannot be readily assessed by the analysis of the circulating pool. Fate mapping studies in mice have suggested that T_{RM} cells have some plasticity and may gain access to the circulating pool (Behr et al., 2020). However, once circulating, these "ex" T_{RM} cells bear most features of other circulating effector memory cells and importantly have no discriminating surface markers. In humans, CD103 (of the canonical T_{RM} cell markers)-expressing cells can be found within both $CD4^+$ and $CD8^+$ blood T cell subsets; however, mRNA analysis revealed no solid evidence for a former tissue residency of these cells (Hombrink et al., 2016). Thus, as "liquid biopsies" of the resident immune pool are challenging if not impossible, decision making for clinical immunologists will in part remain dependent on immune cells that have to be obtained from more invasive and discomforting procedures, e.g., BAL fluids, induced sputum, or fine-needle aspirates. Still, as technological advances now allow detailed molecular analyses of low cell numbers, including monitoring donor T_{RM} cell persistence and recipient T_{RM} cell replacement, the information obtained from these analyses does provide unique insights for diagnosis and treatment.

Glucocortocoids (GCs) are being widely used to dampen immune responses in inflammatory diseases and also reduce recipient immune responses after allogeneic transplantation. GCs have pleiotropic effects on immune cells, including developing and effector-type cells and innate cells and adaptive lymphocytes. As it is well documented that GCs can induce apoptosis of thymocytes (Lépine at al., 2011) but not mature T cells, it is not surprising that the high-dose methylprednisolone the patients received did not lead to an elimination of recipient-derived lung T_{RM} cells. When looking at the development of effector T cells, multiple effects on cytokine production have been documented (Taves and Ashwell, 2021), but evidence of the actions of GCs on gene transcription of cytolytic mediators is sparse (Wargnier et al., 1998). Corticosteroids have broad effects on cellular metabolism and subsequently, as the induction of effector functions is dependent on metabolic pathways (Chang et al., 2013), this might explain at least a part of the beneficial effects of GCs on the allograft-reactive T_{RM} cells. It remains to be investigated if methylprednisolone truly "reprograms" recipient T_{RM} cells, via, for instance, inducing epigenetic alterations in loci that code for effector molecules, or rather silence gene transcription. The finding that T_{RM} cells persist leaves the transplanted lung in a peat fire that may well flare up when the local situation changes as result of, for instance, a viral or bacterial infection. Hence, in line with the narrative of *Inside Job*, GCs may offer temporary local T_{RM} cell regulation, but the underlying system has not changed, and the incentives for allo-antigen–induced immunopathology remain in place.

Lung transplantation is a treatment option for advanced-stage lung disease and survival and improves survival and quality of life in patients (Swaminathan et al., 2021). Although in the past years advances in the diagnosis and the understanding of the pathophysiology of the frequent post-transplant complications have been booked, immune-induced damage, such as ACR and chronic lung allograft dysfunction (CLAD) remains challenging. Two CLADs have been defined, bronchiolitis obliterans syndrome (BOS) and restrictive allograft syndrome. BOS is typified by obliterative bronchiolitis lesions and an obliteration of the small airways, but no prominent pathology is apparent within the parenchyma. Therefore Snyder et al. (2022) rightfully conclude "that clonally expanded $CD8^+$ T cells found during ACR persist as T_{RM} and migrate to the airways suggests a plausible biologic mechanism whereby ACR contributes to BOS."

Although GCs are relatively effective in combination with other immunosuppressives to treat ACR, long-term outcome survival of patients after lung transplantation is still rather poor, reaching a median survival of 6.5 yr (Swaminathan et al., 2021). The disappointing prognosis of this iatrogenic immunopathology urges the development of novel therapeutic interventions, and specific targeting of T_{RM} cells should be considered. In healthy tissue, T_{RM} cells function in protection and homeostasis. Although not addressed here, human lung T_{RM} cells isolated from healthy lung but also from non-small cell lung carcinoma (NSCLC) express high levels of immune checkpoint regulators such as PD-1 and TIM3 (Hombrink et al., 2016; Ganesan et al., 2017). The expression of these immune checkpoint regulators serves to keep the balance between immune-mediated protection against invading pathogens and excessive immunopathology. Blocking of these regulators has proved to be beneficial in melanoma and carcinoma such as NSCLC (Morad et al., 2021). It would be worthwhile to explore if checkpoint agonists and/or co-stimulatory ligand blockers could act at the local level, thereby mitigating early and late immune-mediated damage to tissue. In fact, molecular intervention at this level could have broad implication not only for allo-transplant settings but also for immune-mediated diseases that are refractory to current therapies (Sugiura et al., 2022).

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