the fine-tuning of specific effectors. On the other hand, unveiling the interplay of common transcription factors (for example, RUNX1, KLF6 and RFX2) in charge of the initial and late processes allows one to envision subset-specific therapies, which could represent an additional powerful tool to evaluate the nature and severity of the disease. Indeed, subsets with different maturation scores seem to appear at different magnitudes and in different scenarios, from homeostasis to cancer and chronic inflammation. The elucidation of their chromatin landscapes may provide fruitful information regarding their pathophysiological potential. ❐

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Competing interests

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Check for updates

TFH CELLS

Selenium saves ferroptotic T_{FH} cells to fortify the germinal center

Inhibition of ferroptosis via selenium supplementation promotes the survival of follicular helper T cells, boosting the germinal center and antibody response following vaccination in mice and people.

Michelle A. Linterman and Alice E. Denton

Vaccination creates long-lived humoral immunity by inducing the germinal center (GC) response, an intricate immune reaction that culminates in the production of memory B cells and antibody-secreting plasma cells that protect against (re)infection[1]. The GC is absolutely dependent on help from follicular helper T (T_{FH}) cells[2], but we have yet to fully harness the potential of T_{FH} cells to improve the immunogenicity of vaccines[3]. In *Nature Immunology*, Yu and colleagues[4] report that T_{FH} cells die by ferroptosis, an iron-dependent form of cell death that is characterized by the accumulation of lipid reactive oxygen species[5]. By inhibiting ferroptosis, both genetically and via oral selenium supplementation, the authors were able to boost T_{FH} cell numbers, the GC response and antibody titers after vaccination in both mice and humans. This offers proof-of-principle that targeting T_{FH} cells can boost antibody responses following vaccination to support enduring humoral immunity.

GC B cells present antigen fragments on major histocompatibility complex (MHC)II and engage T_{FH} cells in cognate interactions; in turn, T_{FH} cells provide help, in the form of CD40L-dependent costimulation and cytokines, to GC B cells. These T cell–B cell contacts are critical for the survival of GC B cells and their differentiation into memory B cells and plasma cells[1,2]. GC B cells are constantly searching for help, with dozens of encounters with T cells occurring every hour[6]. Yu and colleagues[4] demonstrate that T_{FH} cells pay the ultimate price for the provision of assistance, as repeated antigen presentation from GC B cells increases the death rate of T_{FH} cells. In both humans and mice, T_{FH} cell death is linked with small and damaged mitochondria and high levels of lipid reactive oxygen species, which are characteristic features of ferroptosis. Genetically modified mice that overexpressed the survival factor Bcl2 or that lacked cell-death-promoting caspase-1 and caspase-11 did not have increased T_{FH} cell numbers in vivo, indicating that apoptosis or pyroptosis were not significantly contributing to T_{FH} cell death. Likewise, culture of human tonsillar T_{FH} cells with chemical inhibitors of necrosis and caspases did not enhance cell survival, whereas blocking ferroptosis did, suggesting that ferroptosis is a significant death pathway for T_{FH} cells.

To investigate the role of ferroptosis in vivo, T cell–specific ablation of glutathione peroxidase 4 (GPX4), a selenium-dependent enzyme that inhibits ferroptosis[5], was used to enable this form of cell death to proceed unhindered. A complete lack of GPX4 resulted in a near-complete abolition of T_{FH} cells and the GC reaction, whereas mice with only one functional allele had fewer T_{FH} cells and GC B cells and reduced vaccine-specific antibodies after immunization, highlighting a dose-dependent role for GPX4 in preventing T_{FH} cell death. Chemical inhibition of ferroptosis in vivo enhanced T_{FH} and GC B cell numbers and antibody titers, demonstrating the key role for ferroptosis in T_{FH} cell biology and robust vaccine responses. The activity of GPX4 depends on the incorporation of selenocysteine into the enzyme, and in conditions where this process is impaired, GPX4 expression is reduced and ferroptosis occurs at a higher rate[5]. Administration of selenium has been reported to upregulate GPX4 expression[7], providing a potential

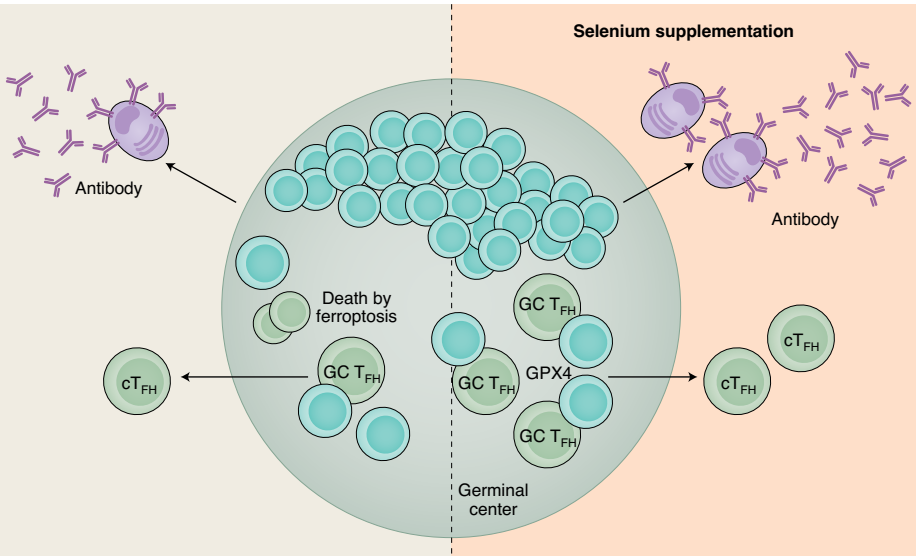

Fig. 1 | Selenium supplementation boosts T_{FH} cells in mice and humans. T_{FH} cells are prone to undergo ferroptosis in the GC, limiting their numbers (left). Yu and colleagues show that supplementing mice and humans with selenium prior to vaccination increases the expression of the selenoenzyme GPX4 and suppresses ferroptosis, resulting in increased numbers of GC T_{FH} cells in mice and their circulating counterpart in humans (cT_{FH} cells) and increased antibody titers following vaccination (right).

therapeutic avenue to limit ferroptosis in T_{FH} cells during vaccination. Oral supplementation of selenium in both mice and humans prior to vaccination increased GPX4 expression, which was linked to higher T_{FH} cell numbers and antibody titers in response to vaccination (Fig. 1). Together, these results demonstrate that dietary selenium supplementation can increase the expression of an antiferroptotic enzyme and enhance vaccine immunogenicity.

This study demonstrates that T_{FH} cells are more sensitive to ferroptosis than other T cell types, including type 1 helper T (T_H1) cells and regulatory T cells. This begs the question of why T_{FH} cells are sensitive to this cell death pathway. Ferroptosis is driven by the accumulation of reactive oxygen species (ROS) that promote lipid peroxidation, which destabilizes the mitochondrial membrane and leads to (ferroptotic) cell death[5]. In the GC, T_{FH} cells are exposed to frequent, repeated TCR triggering and are dependent upon CD28 for their maintenance[2]. Since both TCR and CD28 ligation have been shown to increase ROS in T cells[8], this repeated stimulation may therefore impact T_{FH} cell survival by enhancing the susceptibility of these cells to ferroptosis. In line with this, Yu and colleagues[4] show that T_{FH} cells that are in contact with GC B cells or that have increased antigen exposure have increased intracellular ROS, suggesting a causal link between TCR ligation and production of ROS. The implication of this hypothesis is that the T_{FH} cells with greater TCR signaling will be pushed toward a ferroptotic pathway. TCR signaling drives T cell selection as the GC matures[9]; whether TCR triggering and ferroptosis converge on a common pathway that ultimately drives T_{FH} cell selection during GC development is an interesting concept. What would be the purpose of a ferroptotic rheostat for T_{FH} cells within the GC? Modulation of T_{FH} cells hones the specificity of the GC B cell repertoire and is thought to be important for limiting the development of autoreactive antibodies. Interestingly, P2X7 signaling, which detects extracellular ATP and drives pyroptosis, keeps the T_{FH} cell pool in check in autoimmune settings but not in vaccination[10], suggesting context-specific regulation of T_{FH} cells via cell death.

The identification of a cell death pathway that is more active in T_{FH} cells than in other $CD4^+$ T cell subsets creates an opportunity to intervene in cases where T_{FH} cell formation is known to be impaired, such as in older people after vaccination[11]. The effect of selenium dietary supplementation on vaccine responses was only tested in younger adult mice and people[4], so it is yet to be determined whether this approach will work well in older individuals—arguably the largest sector of society in need of effective vaccines. Strategies for boosting T_{FH} cell responses have typically been limited to changing vaccine adjuvants to enhance either the number or the function of T_{FH} cells[12,13]. Although this is an effective approach for improving humoral immunity following vaccination, the development of new vaccine formulations requires clinical trials, which are time consuming and expensive. By contrast, oral supplementation of a trace element could theoretically be used in conjunction with many different vaccines, which has immediate appeal during a global pandemic and when two vaccine doses are required for optimal efficacy. Approaches to limit ferroptosis may be particularly advantageous for vaccine formats that produce both cellular and humoral immunity, such as messenger RNA–based or adenoviral vector vaccines, as $CD8^+$ T cell expansion and effector function is also reliant on GPX4 (ref. [14]). In addition to being essential for optimal vaccine responses, T_{FH} cells have also been implicated in multiple autoimmune diseases. In such cases, approaches to reduce GPX4 and/or promote ferroptosis may be a potential new therapeutic avenue for targeting T_{FH} cells to limit disease.

The study by Yu and colleagues combines mouse and human immunology to demonstrate that T_{FH} cells are susceptible to ferroptosis. The concordant results between the two species are noteworthy, as they highlight the conserved biology between mouse and human T_{FH} cells and the relevance of using laboratory mice as a preclinical model for the development of vaccine strategies that are relevant to enhancing immunogenicity in humans. One interesting feature of this study is that when ferroptosis is inhibited, it is not just the magnitude of the T_{FH} cell, GC and antibody responses that is improved, but the affinity of the antibody response to the immunizing antigen also increases. This suggests that by preventing the normal processes of T_{FH} cell death, the mechanism of positive selection is altered within the GC. This prompts intriguing questions: do enhanced lipid peroxidation and/or abnormal mitochondria change how T_{FH} cells interact with GC B cells? Has altered expression of B cell helper molecules changed the type of T cell help provided? Further work unravelling how promoting T_{FH} cell survival influences the mutational load, diversity and cross-reactivity of the GC B cell receptor repertoire will provide more insight into how manipulating T_{FH} cell survival influences GC biology and vaccine efficacy to evolving pathogens. ❐

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Competing interests
The authors declare no competing interests.

RESIDENT MEMORY

Resident T cells seek the perfect place to work from home

CD8$^+$ resident memory T (T_{RM}) cells from different tissues form a heterogeneous population. Transforming growth factor (TGF)-β-independent CD103$^-$ T_{RM} cells in the liver retain the ability to move to barrier tissues or return to secondary lymphoid organs.

Hayley A. McNamara and Ian A. Cockburn

Mobility and specialization are two of the key hallmarks of a modern economy. The ability of skilled specialized individuals to come together in vibrant communities creates efficiency and growth. Analogously, the immune system also relies on highly motile cells performing specialized functions in different locations. In this view of the adaptive immune system, secondary lymphoid organs may be seen as bustling cities, where dendritic cells, B cells and T cells gather to find antigen and collaborate to develop robust immune responses. Immune effectors, including CD8$^+$ T cells, then head out to the periphery to control infection. Activated CD8$^+$ T cells adopt a variety of specializations with different migratory patterns. Central memory cells, which remain in the lymphoid tissues, may be seen as metropolitan city dwellers. By contrast, effector memory cells are the commuters of the immune system, shuffling between the lymphoid metropolis and the vast somatic hinterland[1]. More recently, it has been appreciated that populations of CD8$^+$ T cells settle in this hinterland as tissue T_{RM} cells. These are the work-from-home remote employees of the immune system. In this issue of *Nature Immunology*, Christo et al.[2] show that T_{RM} cells—like modern telecommuters—are more diverse and flexible than previously thought (Fig. 1).

T_{RM} cells provide superior protection against pathogens, partly because their location within the tissue makes them the first subset to encounter the pathogen[1]. T_{RM} cells have been identified in both barrier tissues, such as the gut, skin, lung and reproductive tract, and also in the visceral organs, such as the liver[3]. These populations in diverse organs are united by a shared transcriptional program, which is driven by a core group of transcription factors—specifically, by upregulation of Hobit, Blimp-1 and Runx3 and downregulation of Eomes and Tbet[4–6]. The residency of T_{RM} cells has traditionally been established through the use of parabiotic mice, in which the blood supplies of two congenically distinct mice are joined together[1]. In these experiments, tissue-resident populations are predominantly identified within the original animal, displaying minimal capacity to recirculate between animals, unlike their blood-borne counterparts. Analogous methods have also utilized tissue transplantation to show the restricted recirculation of cells from donor tissue into the host circulation[7].

Nonetheless, it has long been clear that there is significant heterogeneity between T_{RM} cell populations. While T_{RM} cells from different organs share a core transcriptional signature, they also express large numbers of organ-specific transcripts[8]. It has been shown that gut T_{RM} cells—even when purified and adoptively transferred—retain a propensity to home to the gut of their new host, suggesting some tissue-specific imprinting of different T_{RM} cells[9]. Cells from different organs are also heterogeneous with respect to their surface markers. In mice, skin T_{RM} cells are almost exclusively CD103$^+$CD69$^+$, whereas liver T_{RM} cells express virtually no CD103. Indeed, liver T_{RM} cells have always been an outlier in the T_{RM} pantheon. Most notably, they do not reside among the parenchymal cells of the liver but, paradoxically, patrol the sinusoidal vasculature and so remain in the bloodstream. Yet these CD69$^+$ liver T_{RM} cells have been shown not to recirculate due to upregulation of the integrin LFA-1, which is critical for their retention in the hepatic sinusoids[10].

In this study, Christo et al.[2] show that dependency on TGF-β is a key driver of the development of phenotypically and functionally distinct resident populations in different organs. TGF-β has previously been found to be crucial for the differentiation of T_{RM} cells within the skin, driving the downregulation of T-bet and the upregulation of the integrin CD103 (ref. [5]).