we injected lampreys with a mixture of plant mitogens and particulate antigens. This immunostimulatory 'cocktail' expanded a population of lymphoblastoid cells, but they, too, did not express signature genes of the adaptive immune system. Their transcriptome was instead enriched in leucine-rich repeat (LRR) sequences (Pancer et al., 2004). Of note, these versatile protein-building blocks are used by every life form on our planet. The Eureka moment came when a summer student's project revealed that each lymphoblastoid cell LRR sequence was different; this remarkable diversity was confirmed by hundreds of sequences, leading us to coin the name 'variable lymphocyte receptors (VLRs)'.

A single germline VLR gene was found by using VLR N-terminal and C-terminal probes to screen a genomic library. It encoded the invariant LRR N and C termini, separated by a non-coding intervening region, plus a terminal stalk region for cell surface attachment (Alder et al., 2005). However, hundreds of potential donor LRR sequences flanking the incomplete germline VLR were found. These provided templates for the stepwise, piecewise assembly of the mature VLRB genes, so called because their products are expressed by B cell-like lymphocytes and plasma cells. Subsequently, VLRA and VLRC were identified and found to be assembled for expression by αβ T cell-like and γδ T cell-like cells, respectively, in both lampreys and hagfish (Bajoghli et al., 2011; Hirano et al., 2013). Therefore, the genetic programme for the separate development of prototypic T cell and B cell lineages must have been present in the last common ancestor of jawed and jawless vertebrates.

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enhanced T_{FH} cell differentiation and germinal centre development, indicating that BCL6 is sufficient to direct T_{FH} cell differentiation. Additional studies found that the majority of IgG production depends on T_{FH} cells.

BLIMP1, a transcription factor encoded by the gene *Prdm1*, was observed to have a reciprocal expression pattern to BCL6. Most T_H1, T_H2 and T_H17 cells express BLIMP1, whereas T_{FH} cells do not and instead express BCL6. Notably, BCL6 represses BLIMP1 expression, and vice versa. This elegant symmetry creates a switch-like bifurcation in CD4$^+$ T cell differentiation, because co-expression of BCL6 and BLIMP1 is a metastable state. *Blimp1*-deficient CD4$^+$ T cells were shown to preferentially differentiate into T_{FH} cells. BLIMP1 overexpression in CD4$^+$ T cells blocked T_{FH} cell differentiation, but allowed for their activation, proliferation and T_H1 cell differentiation in response to viral infection in vivo, providing valuable evidence that BCL6 specifically regulates T_{FH} cell differentiation.

Why did this discovery take 40 years? And why was the recognition of T_{FH} cells so late among CD4$^+$ T cell subsets? In part, there were technical challenges, but the slow progress was more to do with conceptual barriers (the dogma of the T_H1/T_H2 paradigm) and false dead ends along the way, plus some bias regarding initial observations of T_{FH} cells predominantly in human samples. Now, T_{FH} cell biology is a vibrant field, with major impacts on numerous areas of research, including human vaccines, autoimmune diseases, cancers and allergies.

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Journal Club

NRI AT 20: T CELL MEMORY

LYMPHOCYTES IN LOCKDOWN

The field of tissue immunity has undergone a period of explosive growth over the past two decades. Among the many noteworthy studies, it was those by Gebhardt et al. in 2009 and Masopust et al. in 2010 that catapulted research on tissue-resident memory T (T_{RM}) cells into the mainstream. Together, these studies showcased the existence and importance of memory T cells in tissues and the crucial contribution of T_{RM} cells to local immune protection.

Although earlier studies had demonstrated T cell abundance in tissues and supported the notion of their tissue residency (Klonowski et al., 2004; Clark et al., 2006), the studies by Gebhardt and Masopust definitively showed that antigen-specific CD8$^+$ T cells with a unique phenotype persisted in the skin and intestine after local viral infection. Pivotal was the demonstration that these non-migratory T cells were not simply trapped in the tissue, but were a stand-alone population. Moreover, T_{RM} cells mediated site-specific immune responses upon reinfection with the same pathogen.

One of the game-changing outcomes of these studies was the identification of CD69 and CD103 as T_{RM} cell-associated markers (albeit they are not universal T_{RM} cell markers, as shown in later years). This new-found ability to identify T_{RM} cells spurred a flurry of studies that characterized these cells across a range of different tissues in both humans and mice. Subsequent transcriptomic analyses led to insights into how these tissue-embedded cells differ from those in the circulation and revealed the molecular underpinnings of T_{RM} cell development. Most importantly, we have since learnt the value of T_{RM} cells in mediating local immune surveillance, playing a vital role in preventing infection and the development of solid tumours.

Since the Gebhardt and Masopust studies, there has been a seismic shift in how we, as immunologists, study T cell memory. The result is a collective realization of the importance of studying site-specific immune responses and the need to design vaccines that induce tissue-based immunity for effective protection against infections, from influenza to SARS-CoV-2 (Farber, 2021). Understanding how T cells operate in peripheral tissues is key, and by solely looking at blood-borne immunity, we are missing half the story.

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