Kearley, J., Barker, J.E., Robinson, D.S., and Lloyd, C.M. (2005). J. Exp. Med. *202*, 1539–1547.

Lewkowitch, I.P., Herman, N.S., Schleifer, K.W., Dance, M.P., Chen, B.L., Dienger, K.M., Sproles, A.A., Shah, J.S., Köhl, J., Belkaid, Y., and Wills-Karp, M. (2005). J. Exp. Med. *202*, 1549–1561.

Mucida, D., Kutchukhidze, N., Erazo, A., Russo, M., Lafaille, J.J., and Curotto de Lafaille, M.A. (2005). J. Clin. Invest. *115*, 1923–1933.

Umetsu, D.T., and Dekruyff, R.H. (2006). Curr. Opin. Immunol. *18*, 727–732.

Wills-Karp, M. (1999). Annu. Rev. Immunol. *17*, 255–281.

Wilson, M.S., Taylor, M.D., Balic, A., Finney, C.A.M., Lamb, J.R., and Maizels, R.M. (2005). J. Exp. Med. *202*, 1199–1212.

Zhou, L., Lopes, J.E., Chong, M.M., Ivanov, I.I., Min, R., Victora, G.D., Shen, Y., Du, J., Rubtsov, Y.P., Rudensky, A.Y., et al. (2008). Nature *453*, 236–240.

With a Little Help from Their Friends: Interleukin-21, T Cells, and B Cells

Jonathan S. Silver[1] and Christopher A. Hunter[1,*]
[1]Department of Pathobiology, University of Pennsylvania, School of Veterinary Medicine, Room 313, Hill Pavillion, 380 South University Avenue, Philadelphia, PA 19104-4539, USA
*Correspondence: chunter@vet.upenn.edu
DOI 10.1016/j.immuni.2008.06.005

T follicular cells help B cells generate high-affinity antibodies. Two papers in this issue of *Immunity* (Nurieva et al., 2008, and Vogelzang et al., 2008) have identified a role for interleukin-21 in the development of these specialized cells and highlight questions about how this dedicated population is generated.

Since the initial description of the T helper 1 (Th1) and Th2 subsets, the study of T helper cells has been dominated by questions about the events that lead to Th cell differentiation and the role of cytokines and transcription factors that prompt individual T cells to adopt different fates. More recently, with the characterization of regulatory T cells (Treg) and Th17 cells, many of these same issues have been revisited in slightly different contexts. Moreover, with the advent of improved ways to assay polyfunctionality in T cells, it is becoming apparent that defining a T cell lineage based on the production of a single cytokine or expression of a particular transcription factor has its limitations. This in turn has led to debate about whether the paradigm of T cell subsets is still useful and how to characterize phenotypic plasticity within T cell populations. Neglected by many in the midst of these deliberations is the original reason that $CD4^+$ T cells were ascribed a helper function: their ability to provide cognate help to B cells required for the generation of high-affinity antibodies and memory. These interactions between T and B cells provide an important checkpoint for the development of protective antibody responses and are also critical for the maintenance of peripheral B cell tolerance. However, given the distinct compartmentalization of T and B cells within secondary lymphoid tissues, there needs to be a process that brings relevant lymphocytes together. Thus, antigen-stimulated B cells accumulate at the margins of the B cell areas where they elicit T cell help. This carefully choreographed event is dependent on the ability of T cells to provide costimulation to the B cells that leads to the seeding of follicles with B and T cells and lymphocyte proliferation accompanied by differentiation and somatic hypermutation to give rise to plasma cells secreting high-affinity antibodies. The migration of Th cells to the edge of the B cell zones and the follicular regions and germinal centers rich in the chemokine CXCL13 is linked to the expression of the chemokine receptor CXCR5 (Ansel et al., 2000).

Until relatively recently, there has been a poor understanding of the Th cells that are involved in this process, and one model was that Th1 or Th2 cells provided help to B cells through shared mechanisms that include costimulatory interactions through CD40-CD40L or ICOS-ICOSL or their ability to produce specific B cell growth factors. Consistent with this notion was the association of their signature cytokines IFN-γ and IL-4 with class switching and particular IgG isotypes. In contrast, the first transcriptional profiling of $CXCR5^+$ $CD4^+$ T cells was more in line with the idea that T follicular helper (Tfh) cells were a distinct subset (Chtanova et al., 2004). Indeed, from these studies came the ideas that Tfh cells make IL-10 and IL-21, which are not typically associated with Th1 or Th2 cells, but which provide proliferative signals to B cells.

In the last month, three manuscripts have addressed the relationship of Tfh cells with other T cell subsets and identify IL-21 as a key cytokine that promotes the development of these specialized effector cells (Nurieva et al., 2008; Suto et al., 2008; Vogelzang et al., 2008). IL-21 is closely related to other potent T cell growth factors, such as IL-2 and IL-15, and signals through the shared common γ chain. Previous studies had shown that B cells express the IL-21R, that IL-21 promotes growth of mature B cells, and that *Il21r*$^{-/-}$ mice had reduced B cell responses (Spolski and Leonard, 2008). Together, these studies were congruent with the idea that Tfh cells are a source of IL-21 that drives B cell expansion within germinal centers. However, these more recent publications have

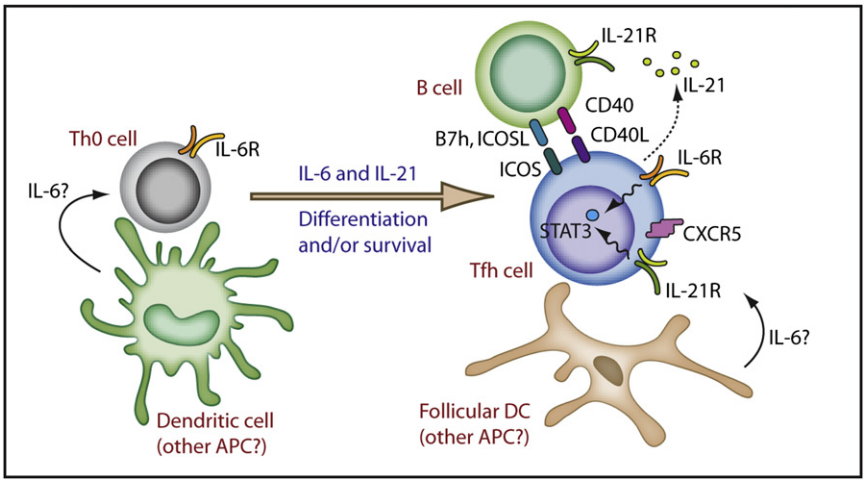

Figure 1. Development and Function of Tfh
Naive Th (Th0) cells presumably activated by dendritic cells and under the influence of IL-6 and/or IL-21 give rise to Tfh that express CXCR5 as well as the IL-21R. The activation of STAT3 is required for the expansion of these cells and at this point, they can provide help to B cells through various costimulatory molecules (CD40-CD40L and ICOS-ICOSL) to promote class switching and affinity maturation. The role of different antigen-presenting cells (APC) in the process of expanding and maintaining the Tfh cells is uncertain.

revealed that IL-21- or IL-21R-deficient mice have a severe defect in numbers of Tfh cells, an observation suggesting either that B cells influence these cells or that IL-21 is a growth factor for Tfh cells (Nurieva et al., 2008; Vogelzang et al., 2008). Somewhat unexpectedly, Tfh cells express IL-21R and IL-21 promotes their expression of CXCR5 (Vogelzang et al., 2008). Thus, it now appears that while IL-21 promotes B cell proliferation there is also an autocrine loop in which T cell-derived IL-21 promote expression of CXCR5, which directs these cells to interact with B cells in the follicular regions and germinal centers (Figure 1).

The identification of IL-21 as a key player in the generation of Tfh cells has led to a new series of questions about the biology of these events. One of these relates to dissecting the ability of IL-21 to promote CXCR5 expression, which would drive T cells to interact with B cells versus its potential to induce proliferation and differentiation of Tfh cells. A related question is which antigen-presenting cells do the Tfh cells interact with at different points in their activation (see Figure 1). Are conventional DC involved in the initial priming of these cells or are B cells and follicular dendritic cells also implicated in these events? Vogelzang et al. (2008) also linked IL-21 to the modulation of TCR mediated signals, and this relates to whether TCR ligation alone is sufficient to start this autocrine loop. Alternatively, there may be an innate source of IL-21 that initiates these events or other environmental cues could induce production of IL-21. One likely candidate is IL-6, which is secreted by DC and FDC and which can promote Th17 cell production of IL-21 (Zhou et al., 2007). Indeed, two of the recent studies have concluded that Tfh cells develop preferentially in an IL-6-rich environment (Suto et al., 2008) and showed that *Il6*$^{-/-}$ mice had fewer Tfh cells (Nurieva et al., 2008). However, the basis for this latter finding remains unexplored but provides an attractive explanation for the B cell defects in the absence of IL-6.

Pertinent to the studies described above, Hsu et al. (2008) recently presented evidence that IL-17 would drive an autoimmune process by promoting the formation of spontaneous germinal centers in a model of arthritis associated with high levels of circulating immune complexes. Although there are many possible interpretations of these data, when combined with the ability of IL-21 to promote Th17 cell activity (Korn et al., 2007; Zhou et al., 2007), it is timely that all three of the most recent studies also start to assess the relationship of Tfh with Th17 cells. This issue has been experimentally addressed by Nurieva et al. (2008) whose gene-profiling approach showed that Tfh cells shared some common transcriptional signatures with Th17 cells, most notably IL-21 and the transcription factor STAT3, but their development is independent of TGF-β, and the retinoid related orphan receptors RoRα and RoRγ, three factors that contribute to Th17 development. However, the key difference here is that the BXD2 mice used by Hsu et al. (2008) represent a model of active autoimmunity whereas the two reports published here (Nurieva et al., 2008; Vogelzang et al., 2008) are based on the study of nonpathological antibody responses. Thus, it remains a possibility that there could still be a role for IL-17 in the regulation of the T and B interactions in different disease settings. This may also be the prelude to the balkanization of Tfh cells based on their functional responses in different disease or infectious settings.

The last point to consider is whether these insights could lead to new therapies or management strategies for conditions in which dysregulated B cell production of antibody underlies autoimmune disease. Under these circumstances, would IL-21 blockade ameliorate the sustained production of pathogenic auto-antibodies in conditions like SLE? Alternatively, because the efficacy of most vaccines appear to be dependent on the amount of circulating high-affinity antibody, understanding how Tfh cells are controlled could provide additional opportunities to optimize vaccine strategies. There has been an emphasis placed on the role of IL-21 in promoting Tfh cell activity, and this in turn implies that a reduction in IL-21 would help to terminate the B cell response and that these events may influence the quality of the B cell memory compartment. Indeed, Fazilleau et al. (2007) establish the idea that there are Tfh memory cells, and although this has been linked to depots of cognate antigen that result in continued T cell activation, it raises the possibility of harnessing these long-lived cells. Perhaps the use of novel approaches that target the production of IL-21 may promote Tfh cell function and memory formation and lead to more sustained titers of antibody without the requirement for repeated boosting.

REFERENCES

Ansel, K.M., Ngo, V.N., Hyman, P.L., Luther, S.A., Forster, R., Sedgwick, J.D., Browning, J.L., Lipp, M., and Cyster, J.G. (2000). Nature *406*, 309–314.

Chtanova, T., Tangye, S.G., Newton, R., Frank, N., Hodge, M.R., Rolph, M.S., and Mackay, C.R. (2004). J. Immunol. *173*, 68–78.

Fazilleau, N., Eisenbraun, M.D., Malherbe, L., Ebright, J.N., Pogue-Caley, R.R., McHeyzer-Williams, L.J., and McHeyzer-Williams, M.G. (2007). Nat. Immunol. *8*, 753–761.

Hsu, H.C., Yang, P., Wang, J., Wu, Q., Myers, R., Chen, J., Yi, J., Guentert, T., Tousson, A., Stanus, A.L., et al. (2008). Nat. Immunol. *9*, 166–175.

Korn, T., Bettelli, E., Gao, W., Awasthi, A., Jager, A., Strom, T.B., Oukka, M., and Kuchroo, V.K. (2007). Nature *448*, 484–487.

Nurieva, R., Chung, Y., Hwang, D., Kang, H.S., Ma, L., Yang, X., Wang, Y., Watowich, S., Jetten, A., Tian, Q., and Dong, C. (2008). Immunity *29*, this issue, 138–149.

Spolski, R., and Leonard, W.J. (2008). Annu. Rev. Immunol. *26*, 57–79.

Suto, A., Kashiwakuma, D., Kagami, S., Hirose, K., Watanabe, N., Yokote, K., Saito, Y., Nakayama, T., Grusby, M.J., Iwamoto, I., and Nakajima, H. (2008). J. Exp. Med. *205*, 1369–1379.

Vogelzang, A., McGuire, H.M., Yu, D., Sprent, J., Mackay, C.R., and King, C. (2008). Immunity *29*, this issue, 127–137.

Zhou, L., Ivanov, I.I., Spolski, R., Min, R., Shenderov, K., Egawa, T., Levy, D.E., Leonard, W.J., and Littman, D.R. (2007). Nat. Immunol. *8*, 967–974.

Bottom Up: A Modular View of Immunology

Ena Wang[1] and Francesco M. Marincola[1,*]
[1]Infectious Disease and Immunogenetics Section, Department of Transfusion Medicine, Clinical Center, National Institutes of Health, 9000 Rockville Pike, Bethesda, MD 20892, USA
*Correspondence: fmarincola@mail.cc.nih.gov
DOI 10.1016/j.immuni.2008.07.002

In this issue of *Immunity*, Chaussabel et al. (2008) apply an inductive approach to pathway discovery identifying modular units that govern human immune biology.

> "It is impossible for the human intellect to grasp ideas of absolute continuity of motion. Laws of motion of any kind only become comprehensible to man when he can examine arbitrarily selected units of that motion. But, at the same time, it is this arbitrary division of continuous motion into discontinuous units which gives rise to a large proportion of human error."
>
> —Leo Tolstoy, *War and Peace*, III: 3

It is increasingly appreciated that monothematic, deductive reasoning applied to the testing of individual genes or proteins does not efficiently embrace the complexity of human immune biology (Marincola, 2007; Benoist et al., 2006). Deductive reasoning aims at confirmation of hypotheses by minimizing experimental variables. However, this reasoning when applied to the clinics needs to confront the uncontrollable nature of human biology molded by the heterogeneous genetic background of patients, their phenotypic adaptations to environmental forces, and, in some instances, the rapid evolution of disease dictated by unstable viral or neoplastic genomes. Thus, nonlinear mathematics may better fit the purpose of comprehending the host reaction to a pathogenic insult in its globality (Dalgleish, 1999). Indeed, biology manifests several characteristics of chaotic systems in which repetitions, given a sufficient number of permutations, progressively exfoliate random associations leaving a bare stem of recurrent patterns linked by necessity to a particular phenomenon. Identification of these recurrent themes segregates relevant from irrelevant observations. Thus, as an alternative to deductive reasoning, inductive reasoning moves from observation to broader generalizations, allowing the formulation of evidence-based hypotheses. This reasoning is the impetus of the work by Chaussabel et al. (2008) in which they applied inductive reasoning to pathway discovery and identified operational units comprising sets of functionally related genes that are differentially expressed by peripheral blood mononuclear cells (PBMCs) in distinct immune pathologies. This discovery provides a framework for an evidence-based approach to system immunology.

Inductive reasoning applied to immunology has to confront the daunting number of permutations arising from thousand of genes interacting with one another. Current technology allows the accumulation of genome-wide information about coordinate expression patterns. However, the extraordinary volume of data generated is not matched by the capacity of the human brain that is "poorly equipped to handle the multidimensionality that results from these broad analyses" (Benoist et al., 2006). Thus, the promises offered by the "omic" revolution produced fewer results than originally anticipated because, in part, of an unprepared audience of biologists and clinicians whose familiarity with bioinformatics principles is not commensurate to the present needs (Bialek and Botstein, 2004).

Deductive reasoning applied to biology traditionally follows a "top-down" approach: A gene or protein is responsible for a given phenotype through a direct cause-effect relationship. However, because genes and their products interact and modulate each other's expression, this linearity is rarely unambiguous and clusters of genes need to be assembled into molecular pathways to explain biological functions. In turn, integration of experimentally defined pathways constructs virtual networks in which biological interactions are predicted according to theoretical algorithms (Avila-Campillo et al.,