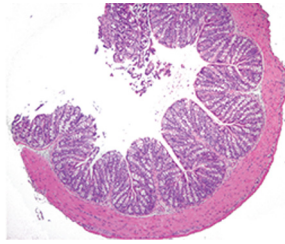


## Righting the SHIP toward Th17

**A**ctivation of the PI3K signaling pathway promotes survival and proliferation of CD4<sup>+</sup> effector Th cells (Teff), whereas inhibition of PI3K is associated with CD4<sup>+</sup>FoxP3<sup>+</sup> regulatory T cell (Treg) differentiation. PI3K regulation is mediated through phosphatases, including Src homology 2 domain-containing inositol polyphosphate phosphatase (SHIP), and Locke et al. (p. 975) have observed that SHIP is a key regulator of Th17 and Treg development. PI3K signaling in Treg and Teff cells was measured by Akt phosphorylation and confirmed earlier findings that phospho-Akt levels were significantly lower in Treg vs Teff cells following TCR stimulation. The mechanism behind reduced PI3K/Akt activation was analyzed by characterizing murine SHIP<sup>-/-</sup> T cells. SHIP deficiency did not alter Treg anergy or suppressive function in vitro, and SHIP<sup>-/-</sup> Treg cells suppressed colitis as effectively as wild-type (WT) Treg in mice. Unexpectedly, SHIP<sup>-/-</sup> Teff cells induced less severe colitis and lower levels of systemic proinflammatory cytokines compared with WT cells. This phenotype was attributed to a defect in Th17 differentiation, possibly due to reduced IL-6-induced STAT3 phosphorylation and an increased tendency toward retinoic acid- and TGF- $\beta$ -driven Treg differentiation. Thus, SHIP appears to be critical for driving Th17 development and balancing Th17 and Treg differentiation.



## The Janus of $\gamma\delta$ T Cells and Allergy

**T**he increasing burden of allergic diseases on human health has drawn attention to studies aimed at understanding the mechanisms behind IgE responses. Previous studies have suggested that  $\gamma\delta$  T cells can both enhance and suppress IgE responses, and Huang et al. (p. 849) show that different  $\gamma\delta$  T cell subsets promote these contradictory responses. IgE responses were examined before or following OVA sensitization in transgenic mice expressing or lacking various TCR genes or in normal mice depleted of different T cell subsets. The results indicated that  $\gamma\delta$  T cells promoted IgE responses in OVA-sensitized mice. IgE responses were enhanced by V $\gamma$ 1-expressing  $\gamma\delta$  T cells (V $\gamma$ 1<sup>+</sup> cells) but suppressed by V $\gamma$ 4-expressing  $\gamma\delta$  T cells (V $\gamma$ 4<sup>+</sup> cells). The suppressive phenotype of V $\gamma$ 4<sup>+</sup> cells included expression of V $\delta$ 5, CD8, and IFN- $\gamma$ . IgE suppression was also observed in naive mice adoptively transferred with V $\gamma$ 4<sup>+</sup> cells isolated from mice sensitized with OVA by inhalation. IgE responses were enhanced by transfer of naive but not OVA-sensitized V $\gamma$ 1<sup>+</sup> cells, suggesting that OVA sensitization affects  $\gamma\delta$  T cell subsets differently. These

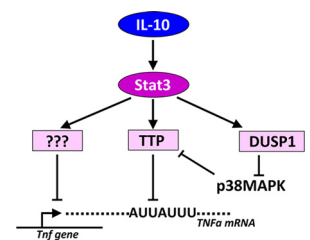
results clarify a role for different types of  $\gamma\delta$  T cells in IgE responses and may bolster future allergy research focused on therapeutic interventions that modulate  $\gamma\delta$  T cells.

## Nuts and Bolts of IFN- $\gamma$ Signaling

**I**nterferon- $\gamma$  induces macrophages to produce inflammatory molecules, including NO, TNF- $\alpha$ , and RANTES. Tsai et al. (p. 856) provide evidence that glycogen synthase kinase-3 $\beta$  (GSK-3 $\beta$ ), a serine/threonine kinase, is a key regulator of these responses. GSK-3 $\beta$  was activated in IFN- $\gamma$ -treated RAW264.7 macrophages through rapid dephosphorylation of Ser<sup>9</sup> by okadaic acid-sensitive protein phosphatases. IFN- $\gamma$  also activated NO production via ceramide generation by neutral sphingomyelinase. In addition, GSK-3 $\beta$  activation was dependent on Tyr<sup>216</sup> phosphorylation by proline-rich tyrosine kinase 2 (Pyk2). Upstream activation of Pyk2 entailed signaling through the phosphatidylcholine-specific phospholipase C (PC-PLC), protein kinase C (PKC), and Src pathways, likely initiated by bioactive lipids. IFN- $\gamma$  receptor (comprised of IFNG1 and IFNG2) engagement activated the Jak-STAT pathway, and interactions between IFNG2 and Jak2 appeared to contribute to PC-PLC, Pyk2, and GSK-3 $\beta$  activation. The mechanisms used by activated GSK-3 $\beta$  to promote inflammation were not elucidated but appeared to involve Src homology-2 domain-containing phosphatase 2 (SHP2) inhibition, thus allowing STAT1-regulated transcription of IFN-inducible genes. These results define GSK-3 $\beta$  as a regulator of IFN- $\gamma$  responses and further our understanding of the signaling events involved in inflammation.

## Tristetraprolin Enacts IL-10's Will

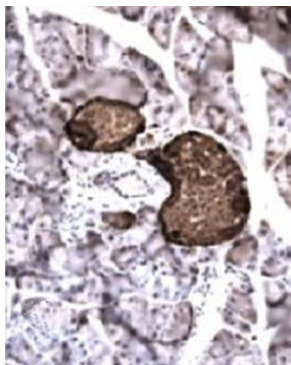
**I**nterleukin-10 is a well-known anti-inflammatory molecule that is vital for turning down the production of proinflammatory cytokines, including TNF- $\alpha$  and IL-1 $\alpha$ . Activation of the Stat3 transcription factor by IL-10 triggers responses involved in regulating proinflammatory cytokines, and Schaljo et al. (p. 1197) have deciphered the multifaceted contributions of the mRNA-destabilizing factor tristetraprolin (TTP) to these responses. TTP was identified previously as a posttranscriptional regulator that degrades proinflammatory cytokine mRNAs with 3' untranslated regions that contain AU-rich elements. In this study, TTP expression was up-regulated in LPS-treated bone-marrow derived macrophages (BMDMs) by IL-10 treatment through a Stat3-dependent mechanism. TTP expression was required for maximal IL-10-induced degradation of TNF- $\alpha$  and IL-1 $\alpha$  transcripts. The ability of IL-10 to increase TTP expression alone was not sufficient to drive mRNA degradation, but IL-10 also suppressed p38MAPK function. Inhibition of



p38MAPK by IL-10 occurred at later times and allowed the subsequent transition of TTP to its active state. These results provide insight into the pivotal role TTP plays in mediating the anti-inflammatory effects of IL-10 in BMDMs.

## Diabetes Delayed through Stem Cells

**L**ittle data exists regarding the efficacy of bone marrow-derived mesenchymal stem cells (MSCs) in animal models of diabetes despite their inclusion in human clinical trials for type 1 diabetes (T1D). Fiorina et al. (p. 993) have bridged this gap by assessing the effectiveness of MSCs in nonobese diabetic (NOD) mice. MSCs isolated from normal BALB/c or NOD mice showed similar phenotypes, except for the higher expression of programmed death ligand 1 on BALB/c MSCs. In vitro, MSCs from both mouse strains were effective at immunosuppressing mixed-lymphocyte reactions and inhibited autoreactive CD4<sup>+</sup> T cells. In contrast, prediabetic NOD mice treated with BALB/c-MSCs, but not NOD-MSCs, had a significant delay in the onset of T1D. Hyperglycemic NOD mice treated with BALB/c-MSCs also had a short-term hyperglycemia reversal, whereas treatment with NOD-MSCs was ineffective. The authors ruled out the influence of mouse strain by showing that MSCs from nonobese resistant (NOR) mice, which are genetically similar to NOD mice, were as effective as BALB/c-MSCs. The authors also observed tumor formation in mice treated with NOD-MSCs, but not with MSCs from BALB/c or NOR mice. These observations support therapeutic development of MSCs for diabetes treatment but also highlight the potential risks associated with this approach.



## Oppression by Opioids

**O**pioids are highly effective analgesics, but chronic opioid use is linked to immunosuppression as well as inhibition of TCR signaling and IL-2 production. Börner et al. (p. 882) have characterized the mechanisms manipulated by opioids to suppress TCR signaling. They confirmed previous reports that IL-2 transcription was inhibited in activated human T cells by pretreatment with morphine or the endogenous opioid beta-endorphin. Opioid suppression was exclusive to cells expressing  $\mu$ - and not  $\delta$ -opioid receptors. Opioids suppressed the immediate signaling events downstream of TCR ligation, including calcium mobilization and phosphorylation of MAPK, the linker of activation of T cells, and ZAP70. Opioid treatment led to an initial decrease followed by a significant rise in cAMP levels in T cells. This rise was linked to activation of protein kinase A and phosphorylation of C-terminal Src kinase (Csk) that in turn preserved phosphorylation of leukocyte-specific protein tyrosine kinase (Lck) at Tyr<sup>505</sup>. Tyr<sup>505</sup> phosphorylation prevented a conformational change in Lck that was essential for its interaction with the TCR and signaling initiation. These results provide detailed evidence that opioids mediate immunosuppression in T cells and suggest

that endogenous opioids may serve as internal regulators of T cell activation.

## Reviving IFN- $\gamma$ 's Antiviral Power

**H**erpes simplex virus-1 infection has been shown to inhibit IFN- $\gamma$  production and this has been linked to HSV-1 activation of suppressor of cytokine signaling 3 (SOCS-3). Frey et al. (p. 1253) showed that HSV-1 induced SOCS-1 expression in a cell type-specific manner and that a SOCS-1 antagonist could inhibit HSV-1 infection. IFN- $\gamma$  pretreatment of HSV-1-infected keratinocytes offered no antiviral protection, although it blocked infection in similarly treated fibroblasts. This difference was linked to a dramatic induction of SOCS-1 gene and protein expression in HSV-1-infected keratinocytes. IFN- $\gamma$ -induced activation of STAT1 $\alpha$  was suppressed by HSV-1 infection in keratinocytes, and SOCS-1 is known to suppress STAT1 $\alpha$  through inhibition of Janus kinase 2 (JAK2). The antiviral activity of IFN- $\gamma$  was restored through reversal of STAT1 $\alpha$  inhibition by keratinocyte treatment with a JAK2 peptide, a SOCS-1 antagonist. Conversely, overexpression of SOCS-1 rendered HSV-1-infected fibroblasts resistant to IFN- $\gamma$ . Keratinocyte infection with an HSV-1 immediate-early protein ICP0 mutant was inhibited by IFN- $\gamma$  and showed reduced SOCS-1 promoter activity, suggesting that ICP0 directly or indirectly altered SOCS-1 expression. These results highlight the role of SOCS-1 during HSV-1 infection and support the development of SOCS-1 antagonists for antiviral therapy.

## Crystal Bares Protease Promiscuity

**T**he lectin pathway of complement is an evolutionarily conserved innate immune response that facilitates recognition of carbohydrate moieties on foreign microorganisms. Carbohydrate structures are bound by mannose-binding lectin (MBL) or ficolins, followed by binding and activation of MBL-associated serine proteases (MASPs) and subsequent complement activation. Dobó et al. (p. 1207) have succeeded in solving the structure of a recombinant catalytic region of MASP-1 (rMASP-1) that includes two complement control protein domains and a serine proteinase (SP) domain. Their observations provide a structural basis for the relative substrate promiscuity of this protease for complement and coagulation component substrates. rMASP-1 showed structural similarities to complement proteins C1s and C1r, as well as MASP-2. However, it contained a novel salt bridge within the primary specificity pocket of the SP domain between the Asp<sup>189</sup> and Arg<sup>224</sup> residues. In addition, the substrate binding region of the SP domain contained a particularly large loop B. Both of these unique features resulted in the SP domain looking more like that of trypsin than of other complement proteases and contributed to the broad specificity of the SP substrate binding groove. These observations afford concrete evidence behind earlier findings regarding the broad substrate specificity of MASP-1 and provide insight into the structural factors that influence protease activity.

