The Journal of Immunology

In This Issue

J Immunol 2013; 190:5907-5908; ;
doi: 10.4049/jimmunol.1390027
http://www.jimmunol.org/content/190/12/5907

Supplementary Material http://www.jimmunol.org/content/suppl/2013/06/10/190.12.5907.DC1

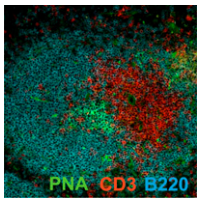
The Journal of Immunology is published twice each month by
The American Association of Immunologists, Inc.,
1451 Rockville Pike, Suite 650, Rockville, MD 20852

Print ISSN: 0022-1767 Online ISSN: 1550-6606.

THE JOURNAL OF IMMUNOLOGY

In This Issue

The Importance of UNG



Uracil-DNA glycosylase (UNG) and the mismatch repair factor MSH2/MSH6 process the deoxyuridine produced when activation-induced deaminase (AID) converts deoxycytidine at the Ig loci. This in turn allows both the class switch recombination (CSR) and somatic hypermutation (SHM) necessary for Ab maturation to occur. Although UNG deficiency reduces CSR but has less of an effect on SHM in vitro, the effects of UNG deficiency have not been tested in vivo. Zahn et al. (p. 5949) have examined the role of UNG in vivo with the use of UNG-deficient ($Ung^{-/-}$) mice. They found that Ab affinity maturation was largely unchanged in $Ung^{-/-}$ mice, although some IgM responses displayed higher affinity compared with wild-type (WT). $Ung^{-/-}$ mice had CSR activity as they had normal or slightly reduced levels of circulating Ig relative to WT and had IgA in the gut as a result of chronic environmental stimulation. However, the responses to acute stimuli were impaired; $Ung^{-/-}$ mice did not produce switched Ig in response to immunization or vesicular stomatitis virus infection. This indicates that the UNG enzyme was necessary for correct CSR during acute but not chronic immune responses.

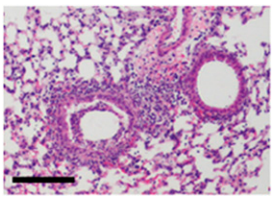
DHMEQ Reacts against NF-κB

Dehydroxymethylepoxyquinomicin (DHMEQ), a potent inhibitor of NF-κB, is effective against various inflammatory and intractable malignant diseases. Reactive oxygen species (ROS) induced by DHMEQ can activate NF-κB; however, sustained ROS production may negatively regulate NF-κB signaling. To evaluate the relationship between DHMEQ-induced ROS and NF-κB, Nakajima et al. (p. 6559) first demonstrated that DHMEQ inhibits TNF-α–induced NF-κB signaling by inhibiting p65 nuclear transport and IKKα/β phosphorylation, preventing degradation of the IκB regulatory protein and subsequent nuclear translocation of NF-κB. Treatment of NRK-52E cells with antioxidants, which block ROS, reversed the DHMEQ-induced block on phosphorylation upstream of the IKK complex. High concentrations of DHMEQ can cause endoplasmic reticulum stress and the authors demonstrated that DHMEQ activated the unfolded protein response (UPR), which could be blocked with antioxidant treatment. Suppression of NF-κB through UPR activation was mediated downstream of the IKK complex through the induction of C/EBPβ. These results demonstrate that DHMEQ inhibits NF-κB through the generation of ROS, which act both upstream of the IKK complex by blocking phosphorylation and downstream of the IKK complex through the activation of the UPR.

microManaging EAE

MicroRNA 155 (miR-155), a small ssRNA molecule expressed in immune cells, has been shown to modulate the development of Th17 cells, which have a significant role in regulating inflammation in autoimmune disorders, such as multiple sclerosis (MS). To investigate the mechanism of miR-155 regulation of Th17 function, Hu et al. (p. 5972) generated miR-$155^{+/+}$ and miR-$155^{-/-}$ IL-17F reporter mice that have one intact allele of the endogenous IL-17F gene. Loss of miR-155 led to defective expression of IL-17F mRNA even under Th17-skewing conditions. Microarray analysis of gene expression in miR-$155^{+/+}$ and miR-$155^{-/-}$ IL-$17F^{+}$ Th17 cells showed a decrease in IL-17, IL-22, and IL-23R and subsequent hyporesponsiveness to IL-23 in miR-$155^{-/-}$IL-$17F^{+}$ Th17 cells. Microarray results also highlighted the Ets1 transcription factor, a known inhibitor of Th17 with a highly conserved binding site for miR-155 in its 3′UTR, and a luciferase assay identified Ets1 as a direct target of miR-155. Ets1 knockdown in miR-$155^{-/-}$IL-$17F^{+}$ Th17 cells upregulated Th17-related genes. The authors generated miR-$155^{+/+}$ and miR-$155^{-/-}$ IL-17F reporter mice with an experimental autoimmune encephalomyelitis (EAE)-inducing transgenic TCR and showed that adoptive transfer of miR-$155^{+/+}$IL-$17F^{+}$Th17 cells into peptide-immunized $Rag1^{-/-}$ mice led to the development of more severe EAE than transfer of miR-$155^{-/-}$ IL-$17F^{+}$Th17 cells. Taken together, the data identify a regulatory pathway for miR-155 involving Ets1 and IL-23 and demonstrate a role for miR-155 regulation of Th17 cells in EAE.

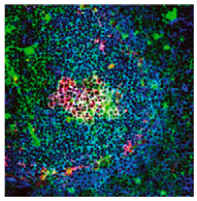
Regulating Virus Responses



Tregulatory cells (Tregs) include "natural" Tregs generated in the thymus based on self-peptide specificity and "adaptive" Tregs that develop from conventional $CD4^{+}$ T cells in the periphery in response to immune signals. During infections, the process by which Tregs develop and how they specialize to acquire new functions and phenotypes are not well understood. Bedoya et al. (p. 6115) examined the role of Tregs in acute influenza virus infection by transferring either thymically-generated $CD4^{+}Foxp3^{+}$ cells or conventional $CD4^{+}Foxp3^{-}$ cells, both expressing an HA-specific TCR, into naive mice. Intrathymically generated Tregs accumulated in the lungs and mediastinal lymph nodes of infected mice, whereas conventional $CD4^{+}$ T cells with the same TCR specificity did not develop into $CD4^{+}Foxp3^{+}$ Tregs. The transfer of virus-reactive Tregs into infected mice led to a significant decrease in effector $CD4^{+}$ and $CD8^{+}$ T cells and a corresponding increase in virus titer. The reduction in effector T cells was mediated by the "specialization" of virus-derived Tregs that differentiated into T-bet^{+} Tregs that secreted IL-10 and

prevented effector T cell proliferation at the site of infection. These data demonstrate that thymically-generated Tregs can be activated by virus-derived peptides and modulate the immune response to virus infection by acquiring a distinct T-bet$^+$ phenotype.

c-Myc AIDs B Cell Differentiation



An effective immune response involves the generation of terminally-differentiated Ab-secreting cells (ASC) and memory B cells. c-Myc, a transcription factor that regulates cell proliferation, differentiation, and apoptosis, is expressed in differentiating and mature activated B lymphocytes; however, its exact role in terminal B cell differentiation is unclear. To evaluate the role of c-Myc in this process, Fernández et al. (p. 6135) used the *c-myc*$^{fl/fl}$ mouse model, which conditionally deleted c-myc. Under in vitro B cell–differentiating conditions, c-Myc–deficient B cells did not proliferate or differentiate into plasmablasts and generated fewer ASC, which hypersecreted IgM but did not produce IgG1 or IgE. Immunization of *c-myc*$^{fl/fl}$ mice with either thymus-dependent (TD) or -independent (TI) Ags impaired splenic ASC generation and led to reduced numbers of marginal zone and follicular B cells compared with control mice. Formation of germinal centers and a normal memory B cell population were impaired in *c-myc*$^{fl/fl}$ mice compared with control mice after TD immunization. Gene expression analysis in specific populations of B cells from *c-myc*$^{fl/fl}$ mice showed an altered transcriptional program with a loss of activation-induced cytidine deaminase (AID; *aicda*) expression, which is required for class switch recombination (CSR). The authors demonstrated that c-Myc transcriptionally regulates AID, thus regulating CSR. Ectopic expression of AID in *c-myc*$^{fl/fl}$ mice restored CSR to IgG1 in c-myc–deficient B cells. Taken together, these data reveal the importance of c-Myc in terminal B cell differentiation and CSR.

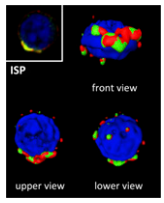
Staying Alive with B7H1

Herpes simplex virus type 1 (HSV-1) causes a latent infection in the trigeminal ganglion (TG) that hovers between acute and chronic disease. When C57BL/6 mice are infected with HSV-1, CD8$^+$ T cells specific for the virus will home to the TG. These TG HSV-1–specific CD8$^+$ T cells exist at a 1:1 ratio, with one population recognizing the HSV glycoprotein B epitope (gB$_{498\text{-}505}$Tet$^+$) and the other recognizing subdominant epitopes (gB-Tet$^-$). Jeon et al. (p. 6277) found that the frequency of gB-Tet$^-$ CD8$^+$ T cells was held constant through higher rates of both proliferation and apoptosis compared with gB$_{498\text{-}505}$Tet$^+$ CD8$^+$ T cells. The higher rate of gB-Tet$^-$ CD8$^+$ T cell apoptosis was consistent with increased expression of programmed death-1 (PD-1) on the TG lymphocytes and B7H1 (PD-1 ligand) on TG neurons. B7H1-deficient mice with latent HSV infection had increased numbers of PD1$^+$ gB-Tet$^-$ CD8$^+$ T cells in the TG compared with wild-type. However, these cells were largely without function and did not provide protection against disease reactivation. Thus, blocking the interaction between PD-1 and B7H1 in latent HSV infection can result in higher numbers of TG CD8$^+$ T cells, but these cells appear "exhausted" and do not function to protect the host.

Xtra CRoss Presenting DCs

The extensive heterogeneity of DC subpopulations has hindered the understanding of the specific roles of DC subsets. To clarify the role of splenic CD8α$^+$ and peripheral CD103$^+$ DCs, which cross-present Ag to generate CD8$^+$ CTL responses and produce high levels of proinflammatory cytokines, Yamazaki et al. (p. 6071) took advantage of XC chemokine receptor 1 (XCR1), which is selectively expressed on these DCs. To create a mouse model with selective ablation of these DC subsets, a fluorescent protein, venus, was knocked into the *Xcr1* locus either alone (XCR1-venus mice), or fused to the diphtheria toxin receptor (DTR) (XCR1-DTRvenus mice), allowing for the ablation of CD8α$^+$ and CD103$^+$ DCs in the presence of diphtheria toxin (DT). DT injection led to rapid but temporary depletion of CD8α$^+$ T cells by day 2 that did not affect other cell populations. Comparison of PBS- or DT-treated XCR1-DTRvenus mice showed that XCR1-expressing cells were: 1) dispensable for in vivo cytokine production after polyinosinic-polycytidylic acid [poly(I:C)] stimulation; 2) essential for CD8$^+$ T cell activation but not CD4$^+$ T cell responses during cross-presentation of OVA protein in the presence or absence of poly(I:C); and 3) crucial for CD8$^+$ T cell responses against cell-associated Ags from dying cells. In this study, the authors demonstrate the critical roles of XCR1-expressing DCs and generate a novel mouse model to study the immune responses of this specific DC subset.

Keeping Wnt under Control



The Wnt signaling pathway is known to play an important role in thymocyte development. Maharzi et al. (p. 6187) have discovered a unique regulator of thymocyte proliferation, transmembrane protein 131-like (TMEM131L), that acts directly on this important signaling pathway. In human thymocytes, TMEM131L expression peaked sharply just before TCRβ-chain selection, leading the authors to hypothesize that it may control fidelity of selection, allowing only thymocytes with successful TCRβ rearrangements to survive. In the absence of TMEM131L, thymocytes hyperproliferated and multiple intrathymic TCRβ-chain selection-related defects were found. TMEM131L-deficient thymocytes also demonstrated constitutive phosphorylation of the Wnt coreceptor LRP6, β-catenin intranuclear accumulation, and higher levels of Wnt target genes compared with wild-type thymocytes. As the name suggests, TMEM131L required association with the cell membrane to be effective. A TMEM131L intracellular isoform showed only marginal inhibition of the Wnt canonical pathway. Membrane-associated TMEM131L controlled Wnt signaling by directing lysosomal degradation of the active, phosphorylated form of LRP6. Thus, TMEM131L is a newly described regulator of thymopoiesis that migrates to the cell membrane before TCRβ-chain selection and controls single positive thymocyte proliferation through the Wnt canonical pathway.