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Integral role of integrins in Th17 development

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Commentary

A lineage of $CD4^+$ T cells known as Th17 cells, which are derived by exposure of naive $CD4^+$ T cells to IL-6 and TGF-β, have been implicated in several autoimmune diseases. In this issue of the *JCI*, studies by Acharya et al. and Melton et al. show that TGF-β is activated at the DC/$CD4^+$ T cell synapse by αv integrins and that this activation is required for Th17 differentiation and autoimmunity in the central nervous system. Thus, these studies offer a potential therapeutic target in fighting autoimmune diseases.

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Integral role of integrins in Th17 development

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A lineage of CD4$^+$ T cells known as Th17 cells, which are derived by exposure of naive CD4$^+$ T cells to IL-6 and TGF-β, have been implicated in several autoimmune diseases. In this issue of the *JCI*, studies by Acharya et al. and Melton et al. show that TGF-β is activated at the DC/CD4$^+$ T cell synapse by αv integrins and that this activation is required for Th17 differentiation and autoimmunity in the central nervous system. Thus, these studies offer a potential therapeutic target in fighting autoimmune diseases.

Th17 cells are a recently identified and critical component of the adaptive immune system (1–3). They are characterized by the production of IL-17A and IL-17F as well as other cytokines such as IL-22. These effector cytokines have been shown to be critical for clearance of certain bacteria and fungal pathogens (4). In addition, vaccine-induced Th17 cells have been shown to have broad protective roles against extracellular pathogens such as *Streptococcus pneumoniae* and to control Th1 cell migration in the context of vaccination against the intracellular pathogen *Mycobacterium tuberculosis* (4). However, this protective aspect of the Th17 lineage comes at a cost, as these cells have been implicated in autoimmune diseases such as multiple sclerosis, psoriasis, and rheumatoid arthritis (1–3).

Several groups have shown that naive CD4$^+$ T cells differentiate into Tregs in the presence of TGF-β (5, 6). However, in the presence of TGF-β and IL-6, naive CD4$^+$ T cells differentiate into Th17 cells (6–8). Early work by Li et al. (9) showed that the source of TGF-β in this context was the CD4$^+$ T cell. However, TGF-β is secreted from cells in an inactive form, in which bioactive TGF-β is in a complex with its latency-associated peptide (LAP) through noncovalent bonds. Two studies in this issue of the *JCI* demonstrate that DCs activate TGF-β in an integrin-dependent fashion (10, 11), suggesting that the activation of TGF-β occurs at the DC/T cell synapse (Figure 1) and that this activation is required to drive the differentiation of Th17 T cells.

TGF-β and integrins

TGF-β is a multifunctional cytokine involved in many aspects of immunology, angiogenesis, and epithelial growth as well as in pathogenic states such as fibrosis (12). Activation of TGF-β has been an area of intense study. Mechanisms identified as leading to the disruption of the noncovalent interaction between LAP and bioactive TGF-β and thus activation of TGF-β

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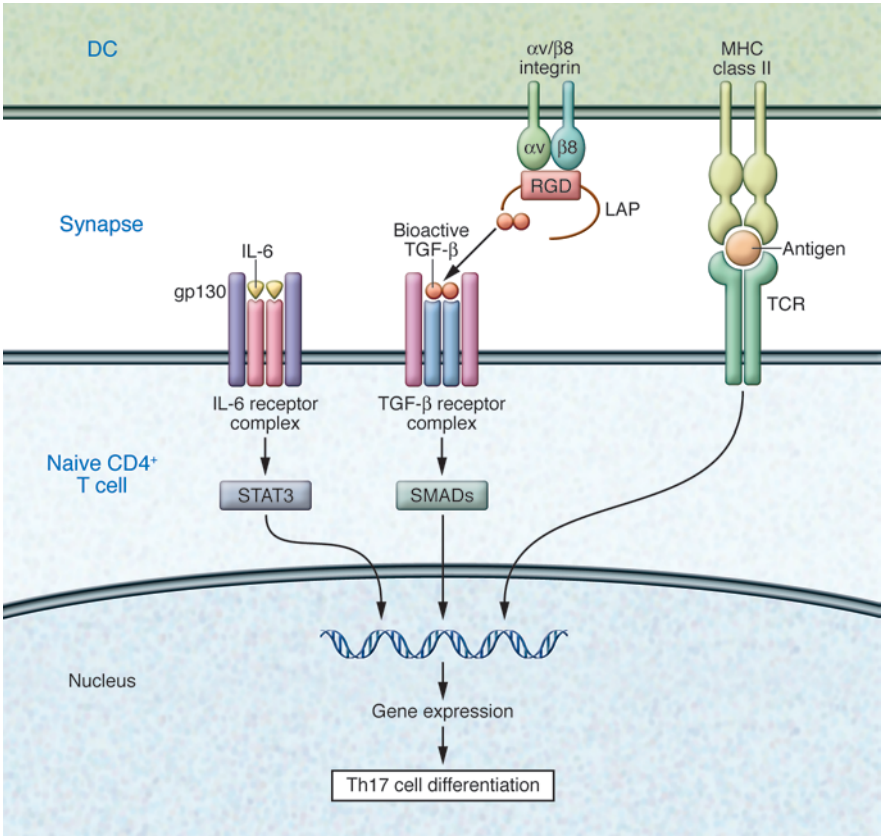

Figure 1
Schematic representation of Th17 differentiation. Two studies in this issue of the *JCI* (10, 11) demonstrate that TGF-β is activated at the DC/CD4+ T cell synapse by αv integrins and that this activation is required for Th17 differentiation in vitro. Moreover, mice lacking αv integrins on DCs fail to develop EAE, a disease mediated by Th17 cells.

include low pH, heat, reactive oxygen species produced as a result of environmental exposures, and LAP cleavage by proteases such as thrombin, elastase, MMP-2, and MMP-9 (13). Because of the ubiquitous expression of TGF-β by many cell types, indiscriminate activation of TGF-β is not advantageous. A more spatially regulated activation occurs through latent TGF-β binding to integrins at the cell surface, which allows activation of TGF-β in a more regulated and localized manner (13).

Integrins are a family of heterodimeric cell surface receptors consisting of an α and a β subunit. There are 24 total integrin subunits (18 α and 6 β). Among the integrins, five share the αv subunit (αvβ1, αvβ3, αvβ5, αvβ6, and αvβ8) and are capable of binding the RGD tripeptide sequence on the LAP of TGF-β (1). Studies in mice that have a mutation converting the RGD sequence of LAP to RGE demonstrate the same embryonic lethality and inflammatory phenotype as mice lacking TGF-β (14), suggesting integrin-mediated activation of TGF-β is critical in development. There are two proposed mechanisms of integrin-dependant activation of TGF-β. In the case of integrins that are bound to the cytoskeleton, such as integrin αvβ6, binding of TGF-β induces a conformational change upon the latent complex of TGF-β, allowing the active portion of TGF-β to be exposed to its receptor, without breaking the LAP/TGF-β bonds (15). Integrin αvβ8 lacks this cytoskeletal connection. In its case, the integrin acts as an anchor for TGF-β, allowing proteolysis by membrane-bound MMP-14 (also known as mt1-MMP) (16). Both of these integrin-related mechanisms allow TGF-β to be activated in a very focal manner, which may be important in the context of Th17 differentiation.

Integrin-mediated Th17 development

In this issue of the *JCI*, two complimentary papers demonstrate the requirement of integrin αvβ8 activation of TGF-β in the differentiation of Th17 cells (10, 11). Previous work using conditional knockout mice has shown that mice lacking either αv (17) or αvβ8 (18) in myeloid cells develop colitis and a spontaneous autoimmune disease, believed to be due to the inability of these mice to activate TGF-β and develop Tregs. Acharya et al. (10) have now considered the common requirement for TGF-β in the development of Tregs and Th17 cells and find that conditional knockout mice (which they generated using tie2-cre and termed αv-tie2 mice) that lack integrin αv on all hematopoietic cells have reduced proportions of Th17 cells in the lamina propria. However, CD4+ T cells from these mice were capable of differentiating into Th17 cells when supplied with exogenous TGF-β in vitro (10). By crossing mice with a floxed *Itgav* allele (i.e., the allele that encodes αv) to LysM-cre mice, which allowed expression of αv integrins on lymphoid cells but not on macrophages and DCs, the authors demonstrated that integrin αv expression on LysM-expressing cells was required for the TGF-β activation that is required for Th17 cell generation in αv-tie2 mice (10). While these data demonstrate the importance of αv, they do not completely identify which integrin is responsible, as mice lacking αv are incapable of making αvβ1, αvβ3, αvβ5, αvβ6, and αvβ8. In support of this work, Melton et al. (11) show a similar phenotype of markedly reduced numbers of Th17 cells in the lamina propria of mice lacking integrin β8 expression on DCs (mice that they term $\beta8^{fl/fl}$ × CD11c-cre mice) (11). Using the experimental model of autoimmune encephalitis (EAE), which is Th17 dependant, neither the αv-tie2 mice (10) nor the $\beta8^{fl/fl}$ × CD11c-cre mice developed EAE (11). To understand what role integrin αvβ8 may have in Th17 development, both groups looked at cytokines involved in Th17 polarization. There were no differences in IL-6, IL-23, TGF-β (10, 11), or IL-1β (10) expression after immunization in the EAE model. Further, IFN-γ, which inhibits Th17 development, was not increased in $\beta8^{fl/fl}$ × CD11c-cre mice, and, thus, the reduced Th17 polarization in vivo could not be explained by enhanced Th1 polarization (11). In vitro, both groups of investigators showed that DCs were required to activate TGF-β, as naive CD4+ T cells did not differentiate in the presence of latent TGF-β unless DCs were present (10, 11). Further, this activation did not occur in the presence of DCs from either αv-tie2 or $\beta8^{fl/fl}$ × CD11c-cre mice or in the presence of RGD mimetics (10) or TGF-β antibodies (11). Interestingly, this activation required cognate interaction

between the CD4+ T cells and DCs (Figure 1), as MHC class II–mismatched DCs, which are unable to present antigen to T cells, did not induce Th17 differentiation (10, 11).

Conclusions

Given the importance of IL-17 in autoimmune disease, the mechanisms of Th17 differentiation are under extensive study. The works presented by Acharya et al. (10) and Melton et al. (11) demonstrate a novel mechanism for the development of Th17 cells, in which naive CD4+ T cells recognize antigens presented by DCs in an MHC class II–dependent manner, while at the same time inducing the cell to differentiate to a Th17 cell through the activation of TGF-β by an integrin αvβ8–dependant mechanism (Figure 1). While these studies do not explain the production of IL-17 by other sources, such as γδ T cells, they do offer insight into the development of an important cell lineage that is implicated in autoimmune states. They also suggest the use of RGD mimetics to block the activation of TGF-β could be a feasible therapy to reduce the severity of Th17-related diseases. Recently, however, work by Ghoreschi et al. demonstrates that Th17 cells can develop in the absence of TGF-β, and Th17 cells grown in these conditions show enhanced pathogenic potential after adoptive transfer (19). These data highlight the complexities of Th17 differentiation and suggest that it will be important to understand the origins and phenotypes of Th17 cells (and their nuanced subsets) in order to develop therapeutic approaches.

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A tincture of hepcidin cures all: the potential for hepcidin therapeutics

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Iron overload as a result of blood transfusions and excessive intestinal iron absorption can be a complication of chronic anemias such as β-thalassemia. Inappropriately low levels of hepcidin, a negative regulator of iron absorption and recycling, underlie the pathophysiology of the intestinal hyperabsorption. In this issue of the *JCI*, Gardenghi et al. demonstrate that increasing hepcidin expression to induce iron deficiency in murine β-thalassemia not only mitigates the iron overload, but also the severity of the anemia. These data illustrate the therapeutic potential of modulating hepcidin expression in diseases associated with altered iron metabolism.

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Hepcidin, iron, and erythropoiesis

Erythropoiesis consumes the majority of the iron present in the human body (1). Most of this iron is obtained from the recycling of effete red blood cells by macrophages found in the liver, spleen, and bone marrow. Interruption of iron export from macrophages leads to functional iron deficiency and iron-limited erythropoiesis. At equilibrium, only a small amount of iron is absorbed in the duodenum from the diet each day. Further, there is no physiologically regulated mechanism of eliminating excess iron from the body. Consequently, the proper regulation of dietary iron absorption as well as iron recycling is essential to maintaining iron homeostasis and to sustaining erythropoiesis.