Interleukin-17—Extended Features of a Key Player in Multiple Sclerosis

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Although multiple sclerosis (MS) is the most common neurological disease of young adults, afflicting hundreds of thousands of people worldwide, its pathogenesis is still only incompletely understood. There seems to be substantial heterogeneity in disease mechanisms, but in the majority of cases an autoimmune origin or at least a decisive autoimmune component is postulated. Therefore, pathogenetic research focuses on different players programmed by the immune system. In this issue of The American Journal of Pathology Tzartos and colleagues extend previous findings about the role of the effector cytokine interleukin (IL)-17 in human disease. IL-17 has recently joined the club of molecules considered as important immunological players for inflammation in the nervous system.

In the past a multitude of studies has addressed the role of the so-called type 1 and type 2 cytokines for adaptive immune responses in MS and its corresponding animal model, experimental autoimmune encephalomyelitis (EAE). The general notion has included that a type 1 response (with CD4 T cells of helper type 1, TH1 cells) represents a proinflammatory, destructive immune reaction whereas a type 2 response (with TH2 cells) reflects a modulatory, nonpathogenic immune reaction and can even protect from autoimmune disease caused by TH1-dependent mechanisms. Study in experimental systems has suggested that TH1 and TH2 cells represent terminally differentiated lineages. Critical effector cytokines in a TH1 response are interferon-γ and tumor necrosis factor-α, which are both implicated in mediating disease pathology in MS and EAE. T-bet, STAT4, and STAT1 are TH1-associated transcription factors. Similarly a TH2 response is characterized by cytokine secretion of IL-4, IL-5, and IL-13 and the transcription factor STAT6.

Because MS and especially EAE were originally considered as TH1-mediated autoimmune diseases, IL-12 secreted by antigen-presenting cells has commonly been implicated as the critical upstream cytokine for the underlying TH1 response, the presence or absence of which determines the character of an evolving immune reaction. This view was later revised when it was realized that indeed IL-12 knockout mice (which lack the specific p35 subunit and not the p40 subunit shared by IL-12 and IL-23) are highly susceptible to EAE induction whereas IL-23 was discovered to be the upstream modulator of the pathogenic signaling pathways. Because IL-23 is crucially involved in the expansion of cells producing IL-17, this view converged with the discovery that IL-17 is produced by TH cells that are distinct from the traditional TH1 and TH2 cell subsets and were thus designated as TH17 cells. Requirements for TH17 cell differentiation were first defined in experimental models (Figure 1). Here, it was shown in parallel by several groups that a combination of IL-6 and transforming growth factor-β is required and that differentiated TH17 cells are maintained and expanded by IL-23, which is unable to drive TH17 differentiation of naïve T cells by itself. Besides IL-17A and IL-17F, these cells produce IL-6, IL-21, tumor necrosis factor-α, and granulocyte macrophage-colony stimulating factor. At the molecular level the transcription factor RORγT (or the human RORC variant 2 ortholog) serves as the master switch. Additionally, STAT3, which is activated by IL-6 and IL-21, acts in concert with RORγT. Very recent data show that the interferon regulatory factor-4 also seems to be critically involved, because Ifnrf1-deficient mice fail to produce IL-17. In contrast, the transcriptional master switch for TH1 cells, T-bet, seems to antagonize TH17 differentiation as does STAT1. The TH1- and TH2-associated STAT4 and STAT6, respectively, are not involved in TH17 differentiation. In the presence of transforming growth factor-β without IL-6 stimulation, there occurs differentiation to regulatory T cells, and it could be convincingly shown that this population originates from the same precursor cells. These cells are able to regulate TH17 cells, as they do for TH1 and TH2 cells, pointing to a reciprocal role...
IL-17 is a crucial effector cytokine with potent proinflammatory effects. It induces the expression of other proinflammatory cytokines such as tumor necrosis factor-α and chemokines, attracts neutrophilic leukocytes, and enhances the maturation of dendritic cells. In the context of acquired immunity, IL-17-producing cells synergize for the control of a variety of bacterial and fungal infections at mucosal surfaces, and IL-17 and other proinflammatory cytokines such as tumor necrosis factor-α and IL-1β cooperate to enhance the expression of antimicrobial peptides. Beyond this invaluable contribution to host defense, IL-17-producing cells are thought to be essential inflammatory mediators in autoimmune diseases such as collagen-induced arthritis, colitis, psoriasis, and EAE. We and others have previously observed that TH17 cells in EAE are CD4 cells and that they are present both in the immune periphery and in the inflamed central nervous system in EAE induced by the MOG-peptide 35-55 in C57BL/6 mice and in PLP-induced EAE in SJL mice. Moreover, neutralization of IL-17 ameliorates clinical disease, a finding that is paralleled by reduced EAE severity in IL-17-deficient animals. It could also be demonstrated that immature TH17 cells exist in the thymus of mice. An interesting new observation is that pertussis toxin, which is widely used as adjuvant for EAE induction in combination with PLP and incomplete Freund’s adjuvant, is able to induce PLP-specific IL-17-producing CD4 T cells in the periphery. Therefore, stimulation of the innate immune system with unrelated environmental pathogens can drive the adaptive immune system toward TH17 differentiation, which in turn can support organ-specific autoimmunity provided that the organism is genetically prone for the development of autoimmunity.

All these experimental analyses concerning IL-17 in mice and EAE focused on CD4 T cells, whereas little is yet known about CD8 T cells. The report in this issue of the *AJP* by Tzartos and colleagues adds some new interesting observations concerning the human disease. A systematic analysis of IL-17-positive cells in the brains of MS patients revealed a significant increase in the number of IL-17+ T cells in the active rather than the inactive areas of MS lesions. Importantly, CD8 T cells were positively immunostained for IL-17 at frequencies similar to those of CD4 T cells in MS tissues. In recent years our view of effector cell populations in MS has been revised. Whereas EAE models with their intrinsic bias toward CD4 T cells have influenced our previous therapeutic strategies, the failure of CD4-directed therapies in MS and the clonal expansion of CD8 T cells in MS lesions and cerebrospinal fluid may identify the true bad gig in the inflamed brain. CD8 T cells came into the focus of MS research when it was demonstrated that CD8+ T cells are more frequent in chronically infected MS plaques compared with CD4+ T cells and that neurons are able to up-regulate MHC I molecules after exposure to interferon-γ, which renders them vulnerable to cytotoxic T-cell attack. Furthermore, axonal damage in MS lesions correlates with the number of CD8 T cells but not with TH subsets. The linkage between CD8 T cells and IL-17 as a crucial effector cytokine, which is described here, adds an important piece of data for future approaches in MS. Also, these findings corroborate previous microarray analyses of MS lesions obtained at autopsy that demonstrated highly increased transcripts of IL-17. Unfortunately, there exist only very few CD8 T-cell-based EAE models at the moment. It would be of great interest to further develop such models to elucidate the contribution of CD8 T cells also in light of the potential role of IL-17 as an effector cytokine in cytotoxic T cells.

Another exciting finding was the fact that IL-17 immunoreactivity could also be detected in astrocytes and oligodendrocytes in active areas of MS plaques. Previously, IL-17 had been found to be expressed in human astrocyte cultures and shown to be up-regulated by tumor necrosis factor-α and IL-1β. Additionally, IL-17-positive astrocytes were found in human and rat brains with acute cerebral ischemia. Therefore, it seems likely that astrocytes (and perhaps oligodendrocytes) can be forced to produce IL-17 under the influence of activating stimuli such as ischemic stress or inflammatory cytokines. This can well be one source, besides infiltrating TH17 cells, for the elevated IL-17 levels found in the cerebrospinal fluid of patients with opticospinal MS. The biological role of this extra T-cell IL-17 remains unclear at the moment, but one can envision a proinflammatory amplification loop resulting in the brain. Future investigations should aim at identifying how such intense inflammation can be terminated, eg, by negative feedback loops.

Because it becomes increasingly clear that IL-17 is an essential player in MS, possibilities for therapeutic intervention are warranted. This, however, does not seem to be an easy task. In the absence of IL-6, FoxP3-positive regulatory T cells are educated, which can modulate inflammation. Indeed, a functional deficit but not a reduction in absolute numbers of regulatory T cells was demonstrated in MS.

**Figure 1.** Molecules involved in the differentiation of murine TH17 cells. Several positive and negative feedback loops are involved, indicated by + and −, respectively. Dashed arrows indicate pathways with limited evidence. See text for details and abbreviations.
patients. \(^{27}\) However, when IL-6 is lacking, as can be achieved in knockout mice, IL-21 can step in and initiate an alternative pathway to induce proinflammatory TH17 cells, as recently shown. \(^{26}\) Indeed, IL-6 seems only to be the first cytokine within a cascade compromising IL-21, which acts in an autocrine amplification loop, and later IL-23, which drives TH17 differentiation (Figure 1). That IL-23 cannot drive TH17 cell differentiation on its own is probably only attributable to the fact that IL-23R is not expressed on naïve T cells. Thus, one can speculate that any signal that up-regulates IL-23R may promote TH17 cell differentiation in an appropriate environment. Therefore, the TH17 cell differentiation pathways get complex, and a potential therapy has to choose carefully an appropriate target. Moreover, there are unexpected species differences, with IL-1β instead of transforming growth factor-β reported to be crucial for TH17 cell differentiation in humans, \(^{29}\) making it more difficult to find adequate therapeutic approaches in animal models. Another unsolved question is the potential interconvertability of TH17 cells and whether there is flexibility in cytokine production, which can be used therapeutically.

More than 2 decades after coining of the TH1/TH2 paradigm, new players have entered the field of autoimmunity. We have learned from previous trials in MS that cytokine networks are often redundant and great care is required when experimental findings from rodent species are translated into human therapy. The findings by Tzartos and colleagues\(^ {1}\) are an important step on this challenging road. They will not only stimulate cytokine-directed therapy but also require specific care when experimental paradigms are transferred to patients.

References


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