Recent Observations on the Heterogeneity of Human Effector Memory T Cells and Novel Cytokines Implicated in Cutaneous Inflammation and Psoriasis

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INTRODUCTION

Psoriasis is a common, chronic skin disease that affects nearly 2.2% of American adults. It carries a substantial burden even when not extensive, and it is a substantial detriment on the quality of life in affected individuals.1–3 The clinical presentation varies greatly: some patients experience limited cutaneous involvement, whereas others have either erythrodermic symptoms that cover large amounts of body surface area or have cutaneous, as well as joint, symptoms.4 Widespread treatment dissatisfaction is common.5,6 Psoriasis traditionally has been thought of as a disorder of epidermal hyperproliferation and decreased epidermal turnover time, resulting in abnormal terminal differentiation and an associated impairment of epidermal barrier function.7,8 Therapeutic approaches, therefore, have used antimetabolites, such as methotrexate or arsenic, to constrain epidermal hyperproliferation.8

With the advent of targeted anticytokine biologic agents, understanding of the pathogenesis of this disease has grown substantially in recent years.9–11 Today, psoriasis is considered by the majority of scientists and clinicians to be an immune-mediated, organ-specific (ie, skin or skin and joints) inflammatory disease. An accepted pathology involves intralesional, skin-infiltrating T lymphocytes that trigger primed basal-stem

ABSTRACT

Psoriasis is a chronic inflammatory skin disease that affects 2.2% of Americans and is a substantial detriment on the quality of life. Psoriasis is a complex interaction between numerous immune cells and has been considered largely a T-helper type 1 (Th1) T-cell–mediated response. Although numerous cytokines, including tumor necrosis factor-αβ, interleukin 1β, and interferon-γ, have been identified with distinct roles in skin inflammation and psoriasis, proinflammatory functions of newly identified cytokines have been linked recently to the alternative T-cell subsets Th17 and Th22 that are distinct in their functions and cytokine profiles. This review summarizes the basic research, translational applications, and new therapeutic developments contributing to the understanding of the diversity of human effector CD4+ Th1, Th17, and Th22 T cells, as well as the proinflammatory cytokines associated with these subsets in psoriasis.
keratinocytes to proliferate and perpetuate the disease process via complex interactions among hyperplastic keratinocytes, dendritic cells (DCs), neutrophils, memory T cells, and macrophages. The immune response in psoriasis traditionally has been considered a T-helper type 1 (Th1)–mediated inflammatory response. During the past few years, emerging evidence suggests the existence of additional, distinct memory T-cell populations, including the Th17 and Th22 subsets, which have been specifically linked to skin inflammation and psoriasis. Numerous cytokines have been identified previously as having important functions in the pathogenesis of psoriasis; these include tumor necrosis factor (TNF)-α, interleukin (IL)-1β, IL-10, IL-12, IL-18, IL-23, and IL-24. However, recent reports illustrate critical proinflammatory and anti-inflammatory roles for newly identified cytokines, including IL-17, IL-19, IL-20, IL-21, IL-22, IL-23A, and IL-24, involved in cutaneous inflammation. The molecular and biologic functions and interactions of these new molecules are still not completely understood within the context of the immunologic circuitry of skin inflammation and disease.

This review summarizes new and exciting developments in the understanding of the diversity of human effector CD4+ T cells in cutaneous inflammation and psoriasis, provides an overview on the roles of cytokines associated with these T-cell subsets, and reviews the current status of the anticytokine therapeutic drug pipeline.

**FUNDAMENTAL PRINCIPLES OF INNATE AND ADAPTIVE IMMUNITY**

The immune system has evolved to protect the human host from infectious insults and tumor cells and to eliminate foreign, potentially toxic chemicals. There are 2 major effector pathways of the human immune system, innate and adaptive immunity, that both contribute to the pathophysiology of psoriasis. The innate immune system, which responds to pathogens almost instantaneously, is the first line of defense against infection and plays a critical role in the initiation of adaptive immunity. Adaptive immunity takes longer to develop and occurs after primary exposure to a pathogen, vaccination, or experimental immunization.

**THE TH1-TH2 PARADIGM: A LANDMARK CONCEPTUAL BREAKTHROUGH**

It has been nearly 26 years since Mosmann et al realized that CD4+ T cells could differentiate from antigen-naive precursors into distinct subsets of effector memory T cells with specialized...
immunoregulatory functions; they described unique T-cell subsets in mice on the basis of their cytokine profiles. By using a panel of antigen-specific mouse helper T-cell clones, Mossmann et al \(^ {64}\) characterized these helper T cells as either type 1 (ie, Th1) or type 2 (ie, Th2). Th1 cells produce IL-2, interferon (IFN)-γ, granulocyte-macrophage colony-stimulating factor, and IL-3 in response to stimulation by antigen with antigen-presenting cells or by concanavalin A. Th2 cells produce IL-3 and IL-4 (ie, B-cell stimulatory factor 1), a mast-cell growth factor distinct from IL-2. \(^ {64}\) IL-3 and IL-4 (ie, B-cell stimulatory factor 1), a mast-cell growth factor distinct from IL-3, and not Th2 T cells, and it is also required for efficient Th2 T-cell production; and induction of IL-24 gene expression. These data have established a transcription factor network that regulates IL-9 and demonstrate how combinations of cytokine signals generate cytokine-secreting potential by altering the expression of a panel of transcription factors. \(^ {72}\)

Several important chronic inflammatory diseases, including multiple sclerosis, type 2 diabetes mellitus, and rheumatoid arthritis, have been referred to as Th1-mediated diseases. \(^ {65}\) It is now well established that Th1 T cells are involved in cell-mediated immunity and host defense against intracellular pathogens. \(^ {66}\) The classic proinflammatory cytokines associated with Th1 T cells include interferon (IFN)-γ, IL-2, TNF-α, and TNF-β (ie, lymphotoxin-α). \(^ {67}\) Th1 T-cell differentiation is promoted by IL-12 and requires the transcription factor T-bet that mediates inheritable modifications of the IFN-γ gene, leading to its expression after antigenic stimulation. \(^ {68}\) Kano et al \(^ {69}\) have shown that the IFN-γ-induced transcription factor interferon regulatory factor-1 (IRF-1) is essential in Th1 differentiation by acting on IL-12rb1, the gene encoding the IL-12 receptor β1 subunit (IL-12Rβ1). Importantly, CD70 was shown recently to be selectively expressed on Th1 T cells and not Th2 T cells, and it is also required for efficient Th1 immunity. \(^ {70}\)

Th2 T cells are involved in humoral immune responses, such as atopic dermatitis, \(^ {3,71}\) allergic diseases, \(^ {72}\) and host defense mechanisms against extracellular parasites. Th2 cell differentiation is promoted by IL-4, -5, -6, -10, and -13; differentiation requires the transcription factor GATA-3. \(^ {3,73}\) Recent publications described a Th2-related T-cell type characterized by the secretion of IL-9 and IL-10. \(^ {74}\) These so-called Th9 cells can differentiate from Th2 T cells in the presence of transforming growth factor-β1 (TGF-β1), or they can differentiate from naive CD4 T cells treated with a combination of IL-4 and TGF-β1. IL-4-activated signal transducer and activator of transcription 6 (STAT6) is required for repressing the expression of T-bet and Foxp3, transcription factors that inhibit IL-9 production; TGF-β1 production; and induction of IRF-4, which promotes Th9 development. \(^ {75}\) Sahoo et al \(^ {76}\) have demonstrated recently that STAT6, along with c-Jun, can facilitate Th2 cell-specific IL-24 gene expression. These data have established a transcription factor network that regulates IL-9 and demonstrate how combinations of cytokine signals generate cytokine-secreting potential by altering the expression of a panel of transcription factors. \(^ {72}\)

### MAINTENANCE OF IMMUNE HOMEOSTASIS

In a process commonly referred to as the classic macrophage activation pathway, \(^ {77}\) macrophages respond to Th1 T cell secretion of IFN-γ by producing TNF-α and toxic forms of oxygen metabolites that destroy facultative intracellular microorganisms residing in cell phagosomes and lysosomes. \(^ {78}\) In the alternative macrophage activation pathway, \(^ {79,80}\) Th2 T-cell production of IL-4 and IL-10 blocks microbial killing mechanisms that are initiated by Th1-derived IFN-γ. \(^ {73}\) A balance between Th1 and Th2 immune responses and cytokine expression profiles is critical to the maintenance of human health and the prevention of autoimmunity. \(^ {59}\) One example includes tissue transplantation, in which Th1 T-cell responses have been implicated in most forms of acute graft of transplant rejection and graft-versus-host disease, whereas Th2 responses have been associated with either protection or chronic rejection. \(^ {81}\)

Immunologists define unique T-cell subset lineages as “cell populations in which a change in cytokine production is promoted by polarizing signals and stably imprinted by a lineage-specifying transcription factor through epigenetic mechanisms.” \(^ {74}\) Although we know about the types of cytokine patterns that human Th cells tend to produce, we understand less about how the patterns themselves are decided. \(^ {82}\) Experimental evidence suggests that the type of antigen-presenting cell that presents antigen to the T cell has a major influence on its profile and that the concentration of antigen presented to the T cell during primary activation influences its choice. \(^ {83,84}\) Because this evidence is derived mostly from mouse models, the actual signals influencing human Th T-cell development are far from being elucidated. Our understanding of the role of specific cytokines in the process is nowhere near complete.
In the years since the *Journal of Immunology* publication by Mossmann et al in 1986, several seminal articles have been published that describe unique subpopulations of CD4+ cells with functions ascribed to suppressor cell function. Until 2005, Th cells could be subdivided into Th1, Th2, or suppressor cells that were shown to secrete IL-10 (eg, Tr1 cells), or TGF-β (eg, Th3 cells). However, these suppressor cells did not have lineage-specific transcription factors associated with them. It was not until the discovery of CD4+/CD25+ regulatory T cells by Sakaguchi et al in 1995 and the subsequent discovery of the transcription factor Foxp3 as a determining factor of these cells that a clear identity could be assigned to this important immune cell type. Notably, another subset of Th cells, now known as follicular Th cells, is critical for B-cell interactions with T cells; these follicular Th cells are identified by the expression of Pax5, a transcription factor that is also essential for B cells. Although the description and explanation of the full function of these particular T cells are beyond the scope of this review, the reader is encouraged to read several excellent recent publications on their development and immunologic functions. Finally, Li et al have shown recently the critical importance of cAMP in the differentiation of Th subsets and their subsequent inflammatory responses. The authors provide evidence that altering cAMP levels in CD4+ T cells could provide an immunomodulatory approach to target specific Th subsets.

**EXPANDING THE TH1-TH2 PARADIGM FOR SKIN INFLAMMATION AND PSORIASIS: TH17 T CELLS, IL-17 PRODUCTION, AND BEYOND**

Within the last 8 years, 2 additional, important T-cell subset lineages involved in skin function and disease have been described (Table 1). Th17 cells primarily produce IL-17 and IL-22, and Th22 cells exclusively produce IL-22. Although the secretion of IL-17 by a particular T-cell subset had been described previously by several groups in the mid-1990s, the designation of Th17 was not accepted until IL-23-induced Th17 T cells were isolated. These T cells were induced from naive lymphocytes independent of the transcription factors STAT1, T-bet, STAT4, or STAT6, and induction required the costimulatory molecules CD28 and ICOS. Subsequently, significant attention has been focused on the human Th17 lineage and on delineation of molecular and cellular mechanisms underlying its role in inflammatory diseases. A number of inflammatory cytokines, including TGF-β, IL-6, IL-21, IL-22, IL-23, and IL-1β, participate in the generation of Th17 cells. The IL-1 family of molecules is also thought to be involved in Th17 and IL-17 regulation. Lee et al showed recently that IL-17 production from cytokine-treated naive human CD4+ T cells was induced by IL-1β and that this induction was blocked by an IL-1R antagonist. These results indicate that human Th17 cell differentiation is regulated via differential expression of IL-1RI, which is controlled by IL-7 and IL-15. Mouse and human Th17 differentiations are both dependent of the transcription factor nuclear hormone receptor retinoic acid–related orphan receptor RORγt; however, the specific cytokines that promote their differentiation have not been determined. Huh et al recently identified the cardiac glycoside digoxin as a specific inhibitor of RORγt transcriptional activity and demonstrated that mouse Th17 cell differentiation was suppressed without affecting differentiation of other T-cell lineages. They additionally observed that digoxin was effective in delaying the onset and reducing the severity of autoimmune disease in mice. Finally, Bovenschen et al showed recently that Foxp3+ regulatory T cells of patients with severe psoriasis have an enhanced propensity to differentiate into IL-17A–producing cells on ex vivo stimulation with IL-23 when compared with the activity of these cells in healthy controls. Other cell types, including natural killer (NK) T cells, lymphoid tissue inducer–like cells, nuclear hormone receptor retinoic acid–related orphan receptor RORC+ NK p46+ cells, and mast cells, secrete IL-17, IL-22, or both. Collectively, a new classification of Th17 T-cell–related human diseases and therapeutic targets continues to emerge.

**Th17 Family of Cytokines**

**IL-17**

IL-17, or IL-17A, is the original member of a group of cytokines called the IL-17 family, which is composed of IL-17A through IL-17F (although IL-17E is known now as IL-25). IL-17F was originally identified as a transcript from a rodent T-cell hybridoma in 1993. IL-17A is a 155 amino acids.
Table 1. Tissue-Signaling Leukocytes Identified to Date

<table>
<thead>
<tr>
<th>Cell Type</th>
<th>Cytokine Secreted</th>
<th>Other Secreted Factors</th>
<th>Evolution</th>
<th>Transcription Factor</th>
<th>Surface Phenotype</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Well characterized in human system</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Th17</td>
<td>IL-17, IL-22</td>
<td>IL-21, IL-26, TNFα (IL-10), CCL20</td>
<td>Differentiation: naive T cell + TGF-β/IL-1β/IL-6; amplification: IL-21; maintenance: IL-23</td>
<td>RORγt, RORα</td>
<td>CD4+CCR4+CCR6+CXCR3+CD161+IL-23R+</td>
<td>Acosta-Rodriguez et al, 200796 Kleinschek et al, 200997 Cosmi et al, 200898</td>
</tr>
<tr>
<td>Th22</td>
<td>IL-22</td>
<td>TNFα (IL-10), FGFs, IL-32γ</td>
<td>Naive T cell + TNFα/IL-6</td>
<td>AhR</td>
<td>CD4+CCR4+CCR6+CCR10+</td>
<td>Trifarì et al, 2009155 Duhem et al, 2009156 Zhang et al, 2011157 Hu and Francis et al, 2012173 Baba et al, 2012160</td>
</tr>
<tr>
<td>NKT</td>
<td>IL-17, IL-22</td>
<td>IFNγ</td>
<td></td>
<td>RORC</td>
<td>CD3+CD56+</td>
<td>Rachitskaya et al, 2008114</td>
</tr>
<tr>
<td>LTi</td>
<td>(IL-17) IL-22</td>
<td>TNFα Lymphotoxin</td>
<td>Unknown (early NK cell?)</td>
<td>RORC</td>
<td>CD3−CD56−NKp44−CD117+CD127+CD161+</td>
<td>Cupedo et al, 2009115</td>
</tr>
<tr>
<td>RORC+ NKp46+</td>
<td>(IL-17) IL-22</td>
<td>TNFα, IFNγ</td>
<td>LtI-like Cells</td>
<td>RORC</td>
<td>CD3−CD56−NKp44−NKp46+NKG2D+CD117+CD127+CD161+</td>
<td>Crellin et al, 2010116</td>
</tr>
<tr>
<td>NK22</td>
<td>IL-22</td>
<td>TNFα Lymphotoxin, IL-26, leukaemia inhibitory factor</td>
<td>LtI-like Cells</td>
<td>RORC</td>
<td>CD3−CD56−NKp44−CD117+CD127+CD141+</td>
<td>Eyerich et al, 2010149</td>
</tr>
<tr>
<td>Mast</td>
<td>IL-17</td>
<td>MCET and NET</td>
<td>IL-23, IL-1β induces degranulation of Mast cells and Neutrophils</td>
<td>Tryptase, Chymase</td>
<td>Tryptase*, Chymase*, MPO*</td>
<td>Res et al, 2010177 Lin et al, 2011118</td>
</tr>
<tr>
<td>Single reports or indirect evidence of existence</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Monocytes and macrophages</td>
<td>IL-17, IL-22</td>
<td>TNFα, IL-6, IL-8</td>
<td>—</td>
<td>RORγt</td>
<td>CD11b+CD68+</td>
<td>Zheng et al, 2007127</td>
</tr>
<tr>
<td>Neutrophil granulocyte (mouse only)</td>
<td>IL-17</td>
<td>—</td>
<td>—</td>
<td>—</td>
<td>—</td>
<td>Eyerich et al, 2010149</td>
</tr>
<tr>
<td>Paneth cells (mouse only)</td>
<td>IL-17</td>
<td>—</td>
<td>—</td>
<td>—</td>
<td>—</td>
<td>Eyerich et al, 2010149</td>
</tr>
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acid protein and is a disulfide-linked, homodimeric, secreted glycoprotein; its molecular mass is 35 kDa. Each subunit of the homodimer is approximately 15 to 20 kDa. All of the IL-17 isoforms are produced as disulfide-lined homodimers, with the exception of the IL-17A/F heterodimer and the noncovalently bound IL-17B homodimer. The structure of IL-17 consists of a signal peptide of 23 amino acids followed by a 123 amino acid chain region characteristic of the IL-17 family. All members of the IL-17 family have a similar protein structure, with 4 highly conserved cysteine residues critical to their 3-dimensional shape, yet they have no sequence similarity with other known cytokines. Phylogenetic analysis reveals that, among IL-17 family members, the IL-17F isoforms 1 and 2 have the highest homology to IL-17A. These 2 isoforms share 55% and 40% amino acid identity, respectively, with IL-17A; the genes encoding both cytokines are located on the same chromosome in both mice and humans. The next highest homology with IL-17A are IL-17B, 29%; IL-17D, 25%; IL-17C, 23%; and IL-17E, 17%. These cytokines are all well conserved in mammals, with as much as 62% to 88% of amino acids conserved between the human and mouse homologs.

IL-17 functions as a proinflammatory cytokine, acting synergistically with TNF-α and IL-1β. It responds to the invasion of the immune system by extracellular pathogens and induces destruction of the pathogen’s cellular matrix, thus increasing chemokine production in various tissues to recruit monocytes and neutrophils to the site of inflammation in a manner similar to IFN-γ. To elicit its functions, IL-17 binds to a type I cell surface receptor called IL-17R, of which there are at least 3 variants, IL-17RA, IL-17RB, and IL-17RC. The receptor for IL-17 is widely distributed; when engaged with its ligands, it induces activation of nuclear factor-κB and Jun amino-terminal kinase signaling pathways in a TNF receptor–associated factor 6–dependent manner. Recently, the immortalized human keratinocyte cell line known as HaCaT cells, incubated with IL-17A, was shown to upregulate the K17 gene and K17 protein expression in a dose-dependent manner. K17, which is an ectopically expressed keratin in psoriatic lesional epidermis, appears to be an attractive target for novel therapies aimed at curtailing psoriasis driven by chronic inflammation induced by TNF-α.
IL-23

Th17 T cells are crucially involved in the pathogenesis of many autoimmune diseases; therefore, understanding their regulation in psoriasis is of high priority. Ichihara et al demonstrated that serum levels of microRNA 1266, a putative regulator of IL-17A, were considerably higher in patients with psoriasis than in healthy controls. However, microRNA 1266 levels showed weak inverse correlations with psoriasis area severity index scores and body surface areas of involved skin. The observation by Aggarwal et al in 2003 that recombinant mouse IL-23 induced the secretion of IL-17 in memory T cells began to elucidate the role of IL-23 in regulating Th17 T cells.

IL-23 is a heterodimeric cytokine composed of 2 subunits; the p40 subunit is shared with IL-12, and p19 is the IL-23α subunit. IL-23, produced by DCs and macrophages, has a diverse array of biologic functions, including mediating inflammatory responses against infection, promoting matrix metalloprotease upregulation, increasing angiogenesis, and reducing CD8+ T-cell infiltration. The IL-23A gene encodes the p19 subunit and the p40 subunit of IL-12B. The receptor of IL-23 is formed by the β1 subunit of IL-12 (ie, IL-12R-β1) and an IL-23-specific subunit, IL-23R. Both IL-23 and IL-12 can activate the transcription activator STAT4 and stimulate the production of IFN-γ.

Knockout mice deficient in p40, in p19, or in either subunit of the IL-23 receptor (ie, IL-23R or IL-12R-β1) develop less severe symptoms of multiple sclerosis and inflammatory bowel disease, and several recent in vitro and in vivo observations support the involvement of the IL-23/IL-17A pathway in the pathogenesis of psoriasis. Mice overexpressing IL-23p19 develop severe inflammation of the skin, and the intradermal injection of IL-23 in murine skin leads to inflammation that more closely resembles the histopathologic features of psoriatic skin than skin inflammation induced by IL-12. Levels of mRNA for the IL-23p19 and common IL-12/IL-23p40 units but not for the IL-12p35 unit are increased in lesional skin of patients with psoriasis; at the protein level, IL-23 is also more abundantly expressed. Furthermore, sequence variation in the genes encoding the common IL-12/IL-23p40 unit and IL-23R is associated with psoriasis.

Somewhat surprisingly, Fitch et al demonstrated that K5.hTGF-β1 transgenic mice, which develop a psoriasis-like disease, had nonelevated levels of IL-23/IL-17 cytokines in lesional skin compared with those in nonlesional and wild-type skin. They concluded that the IL-23/IL-17 inflammatory pathway is not responsible for the maintenance of inflammatory skin disease in this particular animal model.

IL-21

Although Th17 cells are defined by their production of IL-17, they also produce IL-21, IL-22, TNF-α, and IL-6. According to Spolski and Leonard, Th17 cells represent a mixture of cells rather than a homogeneous population for which the specific secreted cytokine profile presumably depends on the cytokine milieu in which the cells develop. IL-17A, IL-17F, IL-22, and IL-21 are all secreted by and involved in the in vivo function of Th17 cells. IL-21 is a type I, 4-α helical bundle cytokine produced by CD4+ T cells after antigen activation. The human IL-21 gene is approximately 8.43 kb and is mapped to chromosome 4; the IL-2 gene is mapped to the same chromosomal location approximately 180 kb away from IL-21. The mRNA product is 616 nucleotides long. IL-21 is expressed in activated human CD4+ T cells and NK cells but not in most other tissues. The IL-21 receptor (IL-21R) is expressed on the surface of T, B, and NK cells. IL-21R is similar in structure to the receptors for other type I cytokines, like IL-2R or IL-15R, and it requires dimerization with the common gamma chain (ie, γc) to bind IL-21. When bound to IL-21, IL-21R acts through the Janus kinase (JAK)/STAT pathway; it uses JAK1, JAK3, and a STAT3 homodimer to activate its target genes.

In addition to its known pleiotropic range of actions, IL-21 was reported recently to play an important role in the generation of Th17 cells after potent IL-21 induction by IL-6. Th17-differentiated cells in vitro expressed much higher levels of IL-21 mRNA and protein; in the absence of IL-6, IL-21 in combination with TGF-β provided an alternative signal for the induction of Th17 cells. In other studies, IL-21 expression was inhibited by TGF-β, which indicated that IL-17 and IL-21 within the Th17-differentiated population are not always generated by the same individual cells. The recent finding that IL-17A induces a positive feedback loop for enhanced IL-6
expression suggests that IL-17 production, in the proper cytokine environment, may be able to amplify IL-21 production.145 Wei et al144 suggested that IL-21 could function as an autocrine growth factor for Th17 cells, and IL-21 was involved in the amplification loop that induces the Th17 pathway.45,126 However, an IL-21-independent Th17 differentiation clearly exists as well.45,145 Intriguing evidence that IL-21 is involved in the pathogenesis of psoriasis was presented by Warren et al.146 In a cohort of patients with early onset psoriasis, they studied 21 genetic variants in 14 genes that are confirmed autoimmune loci and found associations with 2 variants in the IL-2/IL-21 region.146

IL-22

IL-22 was first described in 2000 from murine IL-9–stimulated BW5147 T-lymphoma cells.147 In humans, IL-22 is encoded by the IL-22 gene and is a member of the IL-10 family or IL-10 superfamily, which includes IL-19, IL-20, IL-24, and IL-26.32 IL-22 binds to a receptor that consists of IL-22R and IL-10R2 (ie, IL-10Rβ) units,148 and its binding activates the JAK-STAT pathway (primarily affecting STAT3).149 Through initiation of STAT3 signaling cascades, the cytokine induces proliferative and antiapoptotic pathways, as well as antimicrobial activity, that help prevent tissue damage and aid repair.150 IL-22 was originally thought to be a Th1-associated cytokine but was subsequently found to be most highly expressed by Th17 T cells.151 IL-22 is induced by IL-6, which also promotes the expression of IL-17A and IL-17F48; however, when Th17-differentiated cells were examined at a single-cell level, less than 10% of these cells also expressed IL-22.44 Likely because TGF-β downregulates IL-22 expression.48,127 IL-17A is highly dependent on the transcription factors nuclear hormone receptor retinoic acid–related orphan receptors RORyt and RORα for its expression.152 In contrast, IL-22 expression requires the ligand-dependent transcription factor aryl hydrocarbon receptor (AhR).153 At this time, there are some questions as to the roles of AhR in murine versus human IL-22 expression.154 IL-22 is expressed both by the adaptive arm of the immune system, which includes CD4+ T-cell subsets,155,156 and by innate lymphocytes, which include NK and lymphoid tissue inducer-like cells.157 IL-22 is an important cytokine for the modulation of tissue responses during inflammatory13,154,157,158 and infectious47,157,159 diseases (Figure 3). Baba et al160 demonstrated recently that the AhR ligand VAF347, a small molecular weight compound, induced the development of naive CD4+ T cells to single IL-22–secreting Th22 cells and that it suppressed the generation of T cells secreting either IFN-γ or IL-17 and IFN-γ.160 These investigators concluded that using VAF347 to interfere with AhR functions may provide an efficient way to intervene with autoimmune disease, because it would “enhance the host protective function mediated by IL-22 while preventing the development of Th cells secreting proinflammatory cytokines.”160

Recent studies on new T-cell subsets have confirmed the important roles of T cells in the “immunological” instruction of tissue cells and also demonstrate the important role of feedback regulation for polarization toward distinct T-cell subsets.161 From these findings there appears to be paradoxical effects of Th17 and Th22 cells because their roles in antimicrobial immunity, tissue inflammation, and tissue protection may overlap in function.161 IL-22 has been extensively studied in the skin and has a complex role in the inflammatory processes of psoriasis and other chronic diseases.159,162 Transgenic mice engineered to overexpress IL-22 have an aberrant skin phenotype that resembles psoriasis.46 The IL-22 transgenic pups are born with shiny and stiff skin and die several days after birth. Histological analysis of the skin reveals epidermal thickening and the presence of infiltrating macrophages in the dermal layer.46 Zheng et al127 demonstrated that IL-22 is preferentially produced by Th17 cells and that it mediates acanthosis induced by IL-23, both in vitro and in vivo. Ma et al,163 using the CD4+/CD45 RB (hi)/CD25− scid/scid psoriasis mouse model, observed antimicrobial peptide and proinflammatory cytokine mRNA in skin lesions that were dependent on the p40 subunit common to IL-12 and IL-23. Neutralization of IL-22 prevented the development of disease and reduced acanthosis, inflammatory infiltrates, and the expression of Th17 cytokines. Direct administration of IL-22 into the skin of normal mice induced both antimicrobial peptide and proinflammatory cytokine gene expression.164 Lindroos et al130 demonstrated that IL-23-mediated epidermal hyperplasia in mice requires IL-6 and that IL-6 induces expression of the IL-22R1A receptor. Rizzo et al46 observed that recombinant mouse IL-23 induced ear swelling, epidermal hyperplasia, and IL-17A and IL-22 expression in wild-type mice but not in IL-22 or IL-17A knockout mice. Wang et al164 demonstrated recently that CCR4+/CCR10+ T cells increased in a mouse model of skin irritation and
found that systemic administration of neutralizing antibodies against CCR4 ligands (ie, CCL17 and CCL22) and a CCR10 ligand (ie, CCL27) led to a significant suppression of T-cell migration and skin inflammation. These authors went on to show that the cytokines that were downregulated by the use of the dual antibody treatment were reflective of a Th22 response rather than a Th17 response.163 Finally, Van Bell et al165 showed that, by using the toll-like receptor 7/8 agonist imiquimod (which induces psoriasis-like pathology), the skin lesions observed in wild-type mice were absent in IL-22 knockout mice or in mice treated with blocking anti–IL-22 antibody. Interestingly, a soluble form of IL-22R exists, IL-22 binding protein (IL-22BP).166,167 Although soluble receptors are in principle generated most often through proteolytic cleavage or alternative splicing, IL-22BP is encoded by an independent gene. IL-22BP is mainly expressed by resting DCs and, to a lesser extent, by activated DCs, resting and activated T and B cells, and activated mast cells.167 It binds to IL-22 but not to other members of this cytokine family. In vitro, IL-22BP can inhibit the function of IL-22 and, therefore, is used frequently for in vitro experiments as a naturally occurring antagonist.

**Th22 Cells: An Important, Newly Identified Memory T-Cell Subset Critically Important in Psoriasis**

In 2009, 4 important studies published by international research teams introduced a new memory T-cell subset, Th22 T cells, which appear to be critically involved in skin biology, inflammation, infections, and psoriasis.168 Duhen et al156 initially characterized a population of human skin-homing memory CD4+ T cells that expressed the chemokine receptors CCR10+, CCR6+, and CCR4+; these cells produced IL-22 but not IL-17 or IFN-γ, and the cells had low or undetectable expression of the transcription factors RORγt and T-bet. The differentiation of T cells producing only IL-22 was efficiently induced in naive T cells by plasmacytoid DCs in an IL-6- and TNF-α–dependent manner.156 At the same time, Trifari et al155 described a similar T-cell population distinct from both Th17 and Th1 cells. Downregulation of either AhR or RORγt RNA-mediated interference affected IL-22 production, whereas IL-17 production was affected only by downregulation of RORγt.155 AhR agonists substantially altered the balance of IL-22–producing and IL-17–producing cells.155 Later that year, Fujita et al169 demonstrated that human dermal DCs and Langerhans cells significantly induced IL-22–producing CD4+ and CD8+ T cells from peripheral blood T cells and from naive CD4+ T cells in mixed leukocyte reactions. Importantly, they showed that the majority of IL-22–producing cells induced by Langerhans cells and dermal DCs lacked expression of IL-17, IFN-γ, and IL-4.169 Finally, Eyerich et al170 identified a subset of human Th cells that produced IL-22 and TNF-α (but not IFN-γ, IL-4, or IL-17) that infiltrated the epidermis in individuals with inflammatory skin disorders. These Th22 clones derived from patients with
psoriasis were stable in culture and exhibited a microarray profile quite different from profiles of Th1, Th2, and Th17 cells. These investigators showed that primary human keratinocytes exposed to Th22 supernatants expressed a transcriptome response profile that included genes involved in innate immune pathways and the induction and modulation of adaptive immunity.\(^{170}\) Within the next 2 years, Th17, Th22, and Th1 cells were measured in whole blood obtained from patients with psoriasis; the abundance of these subsets was reduced after treatment with anti–TNF-\(\alpha\) therapy.\(^{39}\) In addition, the increased presence of cytotoxic T (ie, Tc17 and Tc22) CD8\(^+\) T cells in lesional psoriatic skin,\(^{117}\) as well as Tc22 T cells, which also produce IL-22,\(^{171}\) suggests that these types of CD8\(^+\) T cells play a significant role in skin inflammation and the pathogenesis of atopic dermatitis and psoriasis.\(^{1}\) It has been shown recently that the frequencies of both Th22 cells and Th17 cells were elevated in the peripheral blood from patients with ankylosing spondylitis and patients with rheumatoid arthritis.\(^{172}\) Finally, Hu et al\(^{173}\) recently studied cutaneous lymphocyte-associated antigen-positive CD4\(^+\)/CD45RO\(^+\)/CCR4\(^+\)/CCR6\(^+\)/CCR10\(^+\) Th22 T cells obtained from the whole blood of patients with psoriasis who had active disease and who were no longer responsive to soluble p75 TNF-RII fusion protein therapy (etanercept). They demonstrated significantly upregulated human keratinocyte-derived levels of IL-22, IL-32\(\gamma\), TNF-\(\alpha\), and IL-18 but not IL-17A on direct cell-to-cell contact between Th22 cells and human keratinocytes.\(^{172}\) IL-32\(\gamma\) is a NK4 transcript identified in IL-2–activated T lymphocytes and NK cells; it is a newly described proinflammatory cytokine associated with host defense and inflammation in humans.\(^{174,175}\)

**BASIC RESEARCH AND DRUG DISCOVERY RESEARCH TRANSLATED INTO EFFECTIVE ANTICYTOKINE THERAPEUTICS: STATUS OF THE CURRENT PIPELINE**

**IL-23 Pathway Inhibitors**

Treatment with ustekinumab, a fully human monoclonal antibody (mAb) that binds specifically to IL-12/IL-23p40 and neutralizes human IL-12 and IL-23 bioactivity, is a proven effective therapeutic modality for patients with psoriasis,\(^{35,175–177}\) although effects of its long-term use have not been determined.\(^{178}\) The crystal structure of the antigen-binding fragment of mAb (or Fab) for ustekinumab in a complex with human IL-12 has been determined by x-ray crystallography at 3.0 \(\AA\) resolution.\(^{179}\) The x-ray structure and data from subsequent mutational analyses confirm that ustekinumab binds to the same epitopes on p40 in both IL-12 and IL-23 with identical interactions.\(^{177}\) The approval application for another IL-12/IL-23–targeting biologic drug, briakinumab (ABT-874), has been withdrawn in the United States and Europe to allow for additional analyses and clinical trials. The company plans resubmission at a later date.\(^{180}\) Other IL-23 pathway inhibitors in the pipeline include the anti-p19 mAb apilimod (STA-5326), which interfere directly with IL-23 activity; secukinumab (AIN-457); ixekizumab (LY-2439821)\(^{181}\); and brodalumab (AMG-827),\(^{182}\) which target other members of the IL-23 pathway.\(^{175,182}\) Anti–IL-12/IL-23 biologics represent an attractive class of therapeutic targets for the treatment of psoriasis and other immune-mediated diseases; however, additional studies are needed to evaluate the long-term efficacy and safety of these drugs.\(^{174,183}\)

**IL-17/IL-22 Pathway Inhibitors**

Presently, there are 23 different anti–IL-17, anti–IL-17R, and/or anti–IL-22 small molecule–based therapies from biologic or synthetic sources in preclinical or clinical tests by numerous biopharmaceutical or pharmaceutical companies (Table 2).

Although the identification of IL-17 and the Th17 pathway has broadened the potential array of anticytokine modulators that may prove effective as interventions for patients with psoriasis, the current approach to treatment remains unmodified. On average, it takes between 8 and 12 years for a molecule to advance to the clinic and to approval by US Food and Drug Administration; therefore, we anticipate that the discovery and development of these molecules ultimately will result in important clinical applications.\(^{22,184,185}\)

**CONCLUSION**

Th17 and Th22 cells represent new subsets of important human memory T cells that play a role in the pathogenesis of psoriasis. Psoriatic skin contains many DCs and Langerhans cells that are effective at
differentiating these T-cell subsets to produce IL-17, IL-21, IL-22, and IL-23, as well as other recently identified novel cytokines. The next 5 to 10 years will be a very exciting time in which many new biologic therapies will be tested and approved by the US Food and Drug Administration for inflammatory skin diseases. From the advances made since the Th1-Th2 paradigm was originally described, it is clear that targeted, effective, long-lasting, and safer anti–IL-17 or anti–IL-22 pathway therapeutics for patients with psoriasis will soon become available.

**ACKNOWLEDGMENTS**
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Table 2. Selected Anti-Th17 and Anti-Th22 Therapies for Autoimmune Disease

<table>
<thead>
<tr>
<th>Company</th>
<th>Agent</th>
<th>Target</th>
<th>Stage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Centocor</td>
<td>Stelara (ustekinumab)</td>
<td>IL-12 and IL-23 p40 subunit</td>
<td>Approved</td>
</tr>
<tr>
<td>Pfizer</td>
<td>Tofacitinib (CP690550)</td>
<td>JAK kinases</td>
<td>Registration</td>
</tr>
<tr>
<td>Abbot Laboratories</td>
<td>Briakinumab (ABT-874)</td>
<td>IL-12 and IL-23 p40 subunit</td>
<td>Phase 3 Psoriasis additional trials</td>
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<td>Novartis</td>
<td>AIN457</td>
<td>secukinumab)</td>
<td>IL-17A</td>
</tr>
<tr>
<td>Eli Lilly</td>
<td>LY2439821 Mab</td>
<td>IL-17A</td>
<td>Phase 3 Psoriasis and RA</td>
</tr>
<tr>
<td>Amgen</td>
<td>AMG827 Mab</td>
<td>IL-17R</td>
<td>Phase 3??</td>
</tr>
<tr>
<td>Synta Pharmaceuticals</td>
<td>Aplimos</td>
<td>IL-12 and IL-23 agonist</td>
<td>discontinued at Phase 2 RA</td>
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<tr>
<td>Schering-Plough Biopharma</td>
<td>SCH 900222 Mab</td>
<td>IL-23 p19 subunit</td>
<td>Phase 2 Inflammatory diseases **</td>
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<tr>
<td>Eli Lilly</td>
<td>LY2525623 Mab</td>
<td>IL-23</td>
<td>Phase 2</td>
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<tr>
<td>Centocor</td>
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<td>Phase 2</td>
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<td>Ruxolitinib</td>
<td>JAK1 and 2</td>
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<td>Phase 2</td>
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<tr>
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<td>PKC</td>
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<td>IL-17E</td>
<td>Launched Melanoma</td>
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<td>Jansen Biotech</td>
<td></td>
<td>IL-12 and IL-23</td>
<td>Launched Psoriasis and Crohns</td>
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<td>MedImmune</td>
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<td>Discovery RA and SLE</td>
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<td>Discovery autoimmune</td>
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<td>Discovery inflammatory disease</td>
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<tr>
<td>Merck Serono</td>
<td></td>
<td>IL-22 antagonist</td>
<td>Discovery psoriasis and inflammatory disease</td>
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REFERENCES


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