

REVIEW ARTICLE

Th17 cells: new players in asthma pathogenesis

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Abstract

CD4⁺ T effector lymphocytes are distinguished in different subsets on the basis of their patterns of cytokine secretion. Th1 cells, thanks to IFN- γ production, are responsible for cell-mediated immunity against intracellular pathogens, Th2 cells, through the production of IL-4, provide some degree of protection against helminths, and Th17 cells, via IL-17, promote neutrophils recruitment for the clearance of bacteria and fungi. However, beyond their protective role, these T-helper subsets can also be involved in the pathogenesis of several inflammatory diseases. Asthma is an inflammatory disease characterized by different clinical phenotypes. Allergic asthma is the result of an inflammatory process driven by allergen-specific Th2 lymphocytes, whereas Th17 cells are mainly involved in those forms of asthma, where neutrophils more than eosinophils, contribute to the inflammation. The identification in allergic asthma of Th17/Th2 cells, able to produce both IL-4 and IL-17, is in keeping with the observation that different clinical phenotypes can coexist in the same patient. In conclusion, a picture in which different T-cell subpopulations are active in different phase of bronchial asthma is emerging, and the wide spectrum of clinical phenotypes is probably the expression of different cellular characters playing a role in lung inflammation.

The Th1/Th2 paradigm in protection and disease

CD4⁺ T helper (Th) lymphocytes include both effectors (Teff), devoted to protection from pathogens, and regulators (Treg), whose function is to suppress the immune responses toward autoantigens, as well as exogenous antigens when they induce immune responses that can become dangerous for the host. About 20 years ago, two subsets of effector CD4⁺ Th cells with different functions and patterns of cytokine secretion were identified, in both mice and humans, and were named as type 1 Th (Th1) and type 2 Th (Th2), respectively (1, 2). Th1 cells produce interferon (IFN)- γ and are responsible for both phagocyte activation and the production of opsonizing and complement-fixing antibodies, thus being responsible for the protection against intracellular pathogens. On the other hand, Th2 cells produce interleukin (IL) IL-4, IL-5, IL-9, and IL-13 and play some role in the protection against helminths (3). In addition to their protective functions against pathogens, Th1 and Th2 lymphocytes can also contribute to the development of immune-mediated disorders: Th1 cells have been thought to be involved in the pathogenesis of organ-specific autoimmune diseases as well as other chronic inflammatory disorders such as Crohn's disease

(CD), sarcoidosis, and atherosclerosis (4); allergen-specific Th2 cells play a central role in the development of atopic disorders (3). The Th1–Th2 paradigm was maintained until some years ago, when a third subset of CD4⁺ effector Th cells was identified. These effector cells have been named Th17, because the main cytokine they produce are IL-17A and F (5, 6) (Fig. 1).

The discovery of Th17 cells in mice and humans

Although the existence of IL-17 as a product of activated CD4⁺ T cells has been known for more than 10 years, Th17 lymphocytes have only recently been recognized as a distinct subset of Th cells (5, 6). The IL-17 family is composed by five members, designated IL-17A–F. IL-17A is a disulfide-linked homodimeric glycoprotein, consisting of 155 amino acids (7), sharing great homology with IL-17F (55%). Both IL-17A and IL-17F can exist either as IL-17A and IL-17F homodimers or IL-17A–IL-17F heterodimers. The main role of Th17 cells is their ability to recruit and activate neutrophil granulocytes, either directly through IL-8 production (8) or indirectly by inducing the production of colony stimulatory factors (CSF) and CXCL8 (9) by tissue resident cells. More-

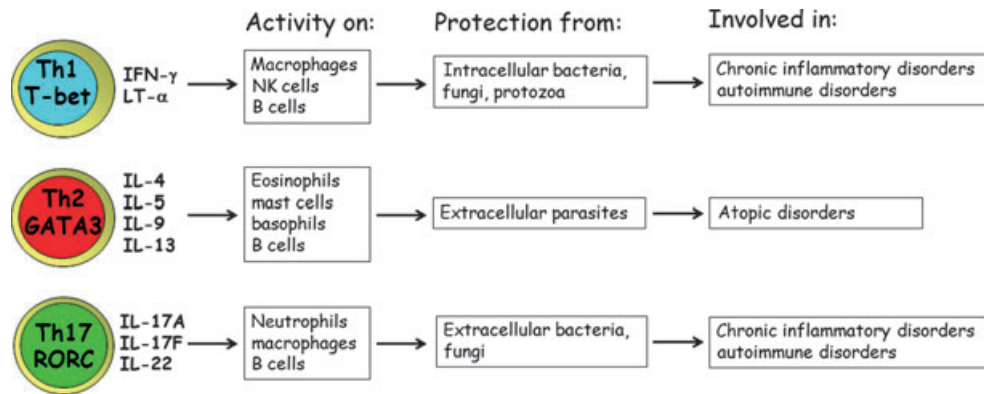


Figure 1 T helper subpopulations: their role in physiology and diseases. Th2 lymphocytes express the transcription factor GATA-3 and produce IL-4, IL-5, IL-9, and IL-13. This subset protects from parasitic infections, but is also involved in the pathogenesis of allergic disease in atopic. Th1 lymphocytes express the transcription factor T-bet and produce IFN- γ and LT- α . They are responsible for the protection from intracellular pathogens and can be involved in

the pathogenesis of chronic inflammatory and autoimmune disorders. Th17 lymphocytes express the transcription factor RORC and produce IL-17A, IL-17F, and IL-22. This population defends the body from infections sustained by fungi and extracellular bacteria, but can also be involved in the pathogenesis of chronic inflammatory and autoimmune disorders.

over, Th17 lymphocytes are also able to stimulate the production of CXCL chemokines and mucins, MUC5AC and MUC5B, in primary human bronchial epithelial cells *in vitro* (10), and the expression of human beta defensin-2 (11) and CCL20 in lung epithelial cells (12). The other IL-17 family members, IL-17B, IL-17C, IL-17D, and IL-17E, are produced by non-T-cell sources. Because of these properties, the main function of Th17 has thought to be the clearance of extracellular pathogens (10, 11).

Human Th17 cells are characterized by the expression of the transcription factor RAR-related orphan receptor C (RORC) (13), by the surface expression of IL-23 receptor (IL-23R), the chemokine receptor CCR6 (14, 15), and the lectin receptor CD161, the orthologous of murine NK1.1 (16, 17). Human Th17 cells originate from CD161+ CD4+ T-cell precursors, detectable in both human umbilical cord blood and thymus, when these cells are activated in presence of a combination of IL-1 β and IL-23 (16). Interestingly, the presence of these cytokines induced not only an increase in the expression of RORC and IL-23R, but also of T-bet and IL-12R β 2 and allowed the development of higher numbers of Th1 cells, suggesting a possible developmental relationship between human Th17 and Th1 cells (16). Moreover, Th17 lymphocytes, showed a lower susceptibility to the anti-proliferative effect of TGF- β than Th1 or Th2 clones (18). This difference appeared to be because of the reduced apoptotic cell death of Th17 cells in presence of TGF- β in comparison with Th1 cells, which appeared to be consistent with the observation that human Th17 cells exhibit lower expression of clusterin (TGF- β signaling, pro-apoptotic), and higher expression of Bcl-2 (anti-apoptotic) in comparison with Th1 or Th2 clones (18). Taken together, these findings support the concept that TGF- β does not have a direct and critical effect on the development of human Th17 cells, but it can indirectly favor their development by selectively inhibiting

both T-bet expression and the development of Th1 cells. Recently, Ghoreschi et al. (19) reported that even murine Th17 cells do not require TGF- β signaling for their differentiation, which occurs in response to the combined activity of IL-1 β , IL-6, and IL-23. In particular, the authors demonstrate, confirming the previous data reported in humans by Santarlasci et al. (18), that TGF- β indirectly promotes the Th17 phenotype by suppressing both T-bet expression and Th1 development (19).

Th17 lymphocytes are not only involved in the clearance of extracellular pathogens during infections (10, 11), but they also play a role in the pathogenesis of several autoimmune and inflammatory diseases (20). The breakthrough leading to the discovery of the Th17 lineage came indeed from murine models of autoimmunity. Experimental autoimmune encephalomyelitis (EAE), collagen-induced arthritis (CIA), and inflammatory bowel disorders (IBD) have been considered for a long time to be the consequence of unchecked Th1 responses (4), on the basis of studies in which disease development was ablated by neutralizing the IL-12p40 chain (IL-12 being a powerful Th1-polarizing agent) or targeting the p40 or the IL-12R β 1 genes. This concept of Th1 association to autoimmune disorders was revised, according to the unexpected discovery that IFN- γ or IFN- γ receptor-deficient mice were more susceptible to central nervous system autoimmunity (21–23). Moreover, a new IL-12 family member, IL-23, was identified in the last years, that shares with IL-12 the p40 subunit, the heterodimer of IL-12 being composed of p40 and p35, and the IL-23 heterodimer being composed of p40 and p19 (5). IL-23 shares with IL-12 also a chain of its receptor, the IL-12R consisting of IL-12R β 1 and IL-12R β 2 chains, and the IL-23R being composed of IL-12R β 1 and the IL-23R chain. Of note, EAE and CIA did not develop in mice deficient in IL-23p19, whereas they could develop in those deficient in IL-12p35 subunit or IL-12R β 2 chain, suggesting that at least in these mod-

els, IL-23 but not IL-12 is critically linked to autoimmunity (6, 24, 25). Based on these data, a determinant role for Th17 cells has been proposed even in the pathogenesis of human autoimmune diseases equivalent to the aforementioned murine models, such as multiple sclerosis, rheumatoid arthritis and IBD, but also psoriasis and contact dermatitis (26, 27). Moreover, the role of Th1 cells, which had been previously shown to be involved in the pathogenesis of the same disorders (3), was underevaluated or even considered as protective against the Th17-mediated inflammation (28). However, after the aforementioned findings obtained in knock out animal models (6, 24), a series of subsequent studies did not confirm the role of Th17, in substitution of Th1, cells as main pathogenic players in chronic inflammatory disorders. In fact, the therapeutic administration of a small interfering RNA specific for T-bet, which limits the differentiation of Th1 lymphocytes, significantly improved the clinical course of established EAE (29). In addition, in *Helicobacter hepaticus*-induced colitis, IFN- γ was the crucial T-cell effector cytokine when T regulatory cells were absent, whereas no effect was reported for IL-17 (30). In proteoglycan-induced arthritis, Th1, but not Th17 cells, appeared to be pathogenic (31). Moreover, either Th1 or Th17 cells were found to be pathogenic in experimental autoimmune uveitis (32). Finally, and more importantly, Th17 cells induced type 1 insulin-dependent diabetes mellitus (IDDM) efficiently in lymphopenic mice only after their conversion into Th1 cells (33). Accordingly, highly purified Th17 cells from BDC2.5NOD mice, completely devoid of IFN- γ at the time of transfer, rapidly acquired the ability to produce IFN- γ in the non obese diabetic/severe combined immunodeficiency (NOD/SCID) recipients, and the development of IDDM was prevented by the treatment with anti-IFN- γ , but not with anti-IL17A-, specific antibody, suggesting that only when Th17 shift toward the Th1 phenotype, became really pathogenic (34). Recently, Kurschus et al. (35), using the IL-17F-Cre^{EYFP} Th17 reporter mouse line demonstrated that both *in vitro* and *in vivo* generated Th17 cells convert into Th17/Th1, or even into pure Th1 cells. Shortly, *in vitro* activated myelin oligodendrocyte glycoprotein-specific enhanced yellow fluorescent protein (EYFP)-positive Th17 cells were transferred into RAG-/- mice. The EYFP-positive cells were recovered from the central nervous system, lymph nodes, and spleen of the mice at the peak of EAE and they were analyzed for IL-17A, IL-17F, and IFN- γ , as well as for Th17- and Th1-specific transcription factors, expression. RT-PCR analysis of EYFP-positive cells recovered from mice at the peak of EAE showed higher IFN- γ and T-bet, and lower IL-17A and IL-17F, mRNA expression than EYFP-positive cells before transfer, while no changes in the expression of RORC and IRF4 were found. The most important point emerging from the study of Kurschus et al. (35) was the definitive demonstration at genetic level that the Th17 response can be quickly lost, as Th17 cells consistently shift to the Th1 phenotype.

A similar conclusion was also obtained in humans; Nistala et al. (36) showed Th17 to Th1 plasticity driven by the inflammatory environment in patients with autoimmune polyarthritis. Likewise, we have found an accumulation of CD4+ CD161+ T cells able to produce IFN- γ (Th1), or

both IFN- γ and IL-17A (Th17/Th1), but not IL-17A alone (Th17), in the synovial fluid (SF) of children with oligoarticular juvenile idiopathic arthritis (JIA) (37). The Th17 cells shared clonal ancestry with CD161+ Th17/Th1 and CD161+ Th1 cells present in the SF, and circulating Th17 cells from healthy subjects demonstrated Th17 to Th1 plasticity *in vitro* in the presence of SF from JIA patients, which could be blocked by IL-12-neutralization. More importantly, the numbers of CD4+ CD161+ cells, particularly the Th17/Th1 cells, in SF of JIA patients positively correlated with levels of erythrocyte sedimentation rate and C-reactive protein (37). Taken together, these findings support the hypothesis that Th17-derived Th17/Th1 and Th1 cells, rather than Th17 cells, play a critical role in disease activity. These findings are of particular importance in light of the debate on the respective role of Th17, Th17/Th1, and Th1 in the pathogenesis of chronic inflammatory disorders and, therefore, in view of developing possible biological therapeutic strategies. With regard to the latter point, it is of note that ustekinumab, a humanized monoclonal antibody that inhibits receptor binding of both IL-12 and IL-23, thus blocking the activity of both cytokines, resulted in a rapid and significant improvement of symptoms in moderate to severe psoriasis, which strongly supports the notion that these cytokines have a key role in the immunopathology of the disease (38, 39). However, the crucial observation supporting a possible involvement of both Th1 and Th17 in the pathogenesis of human autoimmune and inflammatory disorders consists in the fact that these two cell subsets can develop from the same precursors and coexist in the same microenvironment (40) (Fig. 2).

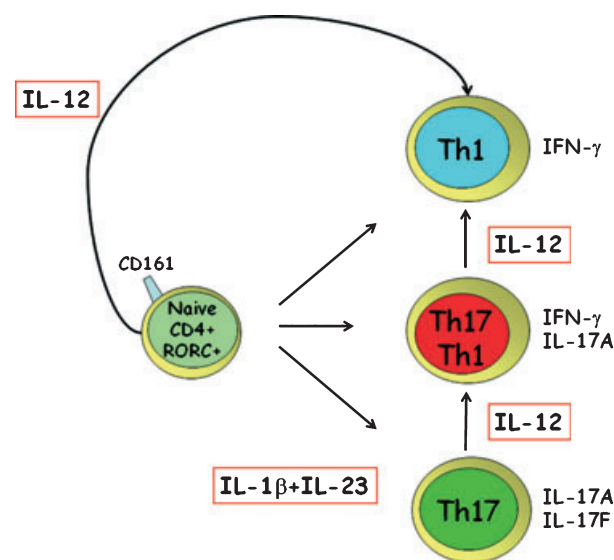


Figure 2 Origin of Th17 lymphocytes and their developmental plasticity toward Th1 phenotype. Human Th17 cells origin from a naive precursor expressing RORC, T-bet, IL-23R, IL-1RI, CD161 and CCR6, exposed to IL-1 and IL-23. In presence of IL-12, the same precursor differentiates toward Th1 phenotype. IL-12 is also able to act on established Th17 lymphocytes driving the transition to Th1, through the intermediate phenotype of Th17/Th1 cells.

Asthma heterogeneity: different phenotypes of disease

Asthma is a heterogeneous inflammatory disorder of the airways characterized by chronic inflammation, airway hyper-reactivity, and by symptoms of recurrent wheezing, coughing, and shortness of breath. Asthma is a major public health problem, affecting 300 million people worldwide, and has increased considerably in prevalence over the past three decades, particularly in Western countries (41). It has been clearly shown that asthma is caused by multiple environmental factors in combination with several major and minor susceptibility genes (42), which can give origin to many different forms or phenotypes of disease. These phenotypes include allergic asthma, for sure the most common form of the disease, severe steroid-resistant asthma (43), asthma induced by air pollutants (44), obesity (45), aspirin (46), and exercise (47). The Global Initiative for Asthma, in 2005, classified asthma according to disease severity, and to the amount of treatment required to achieve control of symptoms and lung function. Alternative methods of classifying asthma are based on the clinical presentation. The clinical presentation is the direct consequence of the nature and the extent of underlying airway inflammation that, for this reason, has to be considered the guide for asthma management (48). The different asthma phenotypes do not necessarily exclude themselves each other, but often coexist and can synergize, although distinct pathogenic mechanisms probably underlie each of them. Th2 immunity is undoubtedly important in supporting bronchial inflammation during allergic asthma being not able to explain the other forms. Many clinical and experimental observations over the past 5 years have suggested that asthma is much more heterogeneous and complex than suggested by the Th2 paradigm. In particular, it has been found that some patients were characterized by the presence of high levels of IFN- γ , IL-17, and neutrophils in the lungs. Notably, these forms of asthma respond poorly to treatment with corticosteroids, the most common therapy for asthma, which seems to benefit mainly patients with allergic asthma. These observations suggest that asthma is indeed heterogeneous, with distinct phenotypes and with different pathogenic mechanisms, some dependent and some independent of Th2 cells, and requiring different therapeutic approaches (41). In this view, the identification of the different inflammatory phenotypes in asthma may allow us not only to better classify the disease, but also to develop new and effective therapies that will be individualized and personalized, taking into account the distinct pathogenic mechanisms occurring in each patient.

The Th2 hypothesis in asthma

Th2 polarization is mainly because of the early production of IL-4 during the primary response (49). However, the cell and the mechanisms responsible for this early IL-4 production remained unclear for a long time. Only recently, it was found that IL-4 could be produced by the naïve Th cell itself, upon Notch triggering, as a consequence of the expression by the dendritic cell (DC) of its ligand Jagged-1 in both mice and

humans (50, 51). Another possibility is the production by other cell types, such as mast cells and macrophages present in the gut of worm-infested animals or lung epithelial cells, of a more recently discovered cytokine, named as IL-25. IL-25 can induce the early production of IL-4 by a non-T, non-B, c-kit⁺, Fc ϵ R1⁺ cell or by the Th naïve cell itself, thus allowing its Th2 polarization (52). Recent studies indicate that not only IL-4, IL-25, and Jagged-1 but also IL-33 and thymic stromal lymphopoietin (TSLP) are important mediators of Th2-cell differentiation (53–55). The role of the type 2 T helper (Th2) cell-mediated immune response against 'innocuous' environmental allergens in the immunopathogenesis of allergic asthma is an established fact. An impressive body of experimental work supports the critical role of cytokines produced by Th2 cells in the initiation, maintenance, and amplification of human allergic inflammation (56). In particular, IL-4 and IL-13 regulate the allergen-specific synthesis of immunoglobulin E, IL-5 the recruitment of eosinophils, IL-9 the growth of mast cells (57). These cytokines also account for other pathophysiological features of both allergy and asthma. IL-4, IL-9, and IL-13 can indeed induce mucus hypersecretion and contribute, together with IL-5, to the increase in airway hyperreactivity (AHR) and, together with TGF- β and IL-6, to airway remodeling. In addition, other mediators, such as prostaglandin and chemokines largely produced in the context of allergic inflammation, can promote the selective recruitment of Th2 cells (58). In this context, it is of note that some chemoattractant receptors are predominantly or selectively expressed by human Th2 lymphocytes (59) and that Th2 cytokines (IL-4, IL-13) are able to increase the production of their ligands. To further underline the role of Th2 lymphocytes in asthma pathogenesis, there is the observation that allergen-specific Th2 cells are present in the lungs of patients with allergic asthma, whereas in chronic obstructive pulmonary disease Th1 lymphocytes have been predominantly found (60, 61). The expression of the allergic phenotype is dependent upon the interaction between genetic predisposition and environmental factors. In this view, the radical changes in lifestyle that have occurred over the past few decades are certainly responsible for the increased prevalence of allergy in Western countries. Several epidemiological studies have clearly shown that modifications of the pattern of microbial exposure of children associated with Westernization represent a critical factor underlying the rising severity and prevalence of atopic disorders (the so-called hygiene hypothesis) (62–64).

An important contribution to the understanding of the role of allergy and Th2 cells in asthma has also benefited from mouse models of allergic asthma. Adoptive transfer of allergen-specific Th2 cells generated from ovalbumin (OVA)-specific T-cell antigen receptor-transgenic DO11.10 mice results in the development of AHR and airway inflammation (65), whereas the transfer of allergen-specific Th1 cells results in a diminution of airway eosinophilia and mucus production (66). These models greatly enhanced the knowledge on the involvement of Th2 cells in allergic inflammation, but reflected only the allergen-driven pathway of asthma. Recent clinical and experimental observations have suggested that

asthma is more heterogeneous and complex than previously thought by the Th2 paradigm and by mouse models of allergic asthma. It has been reported that IL-17 and neutrophils are found in the lungs of patients with asthma, particularly in those with severe asthma or asthma resistant to corticosteroids. In addition, Th2-targeted therapies have not been as effective as hoped in many clinical trials of asthma (41, 67). Furthermore, nonallergic forms of asthma, induced by environmental factors such as air pollutants, viral infection, and obesity, develop independently of Th2 cells (43–47, 68). These observations suggest that asthma is indeed heterogeneous, with distinct phenotypes and with different pathogenic mechanisms operating in different form of disease. In this context, not only Th2 lymphocytes, but also new T-helper subsets, such as Th17 cells, have been proposed to play a role (69) (Fig. 3).

Th17 lymphocytes and asthma

The role of IL-17 in asthma is an area of intense current investigation. In those patients with asthma in which inflammation is nonatopic, non-IgE-dependent, and noneosinophilic, airway neutrophilia is correlated with asthma severity, suggesting a major role for neutrophils, at least in this subset

of patients with asthma (70, 71). Neutrophilic inflammation has also been described in sudden-onset fatal asthma and neutrophil numbers are highly elevated in status asthmaticus (72), thus suggesting a possible role for these cells in severe and fatal asthma. As the role of IL-17 in neutrophil recruitment to the airways is well known, in the last years several studies tried to find an association between Th17 lymphocytes and asthma (73–77). Animal models of asthma suggest that Th17 cells promote neutrophilic inflammation, and, in concert with Th2 cells, are important in the development of airway hyper-responsiveness. Moreover, allergic sensitization through the airway promotes Th17 responses, and IL-17F-deficient mice have an impaired neutrophilic response to allergen (78). More importantly, in humans increased expression of IL-17A and IL-17F has been shown in bronchial submucosa of moderate to severe asthma, and the evaluation of induced sputum in patients with asthma revealed that neutrophilic inflammation was frequently present, particularly in the severe forms of disease (75, 79, 80). Furthermore, it has been reported that increased AHR in response to methacholine in patients with asthma positively correlates with IL-17A levels in the sputum (81). Finally, a polymorphism in IL-17F that results in a loss-of-function mutation is inversely related to asthma risk (82).

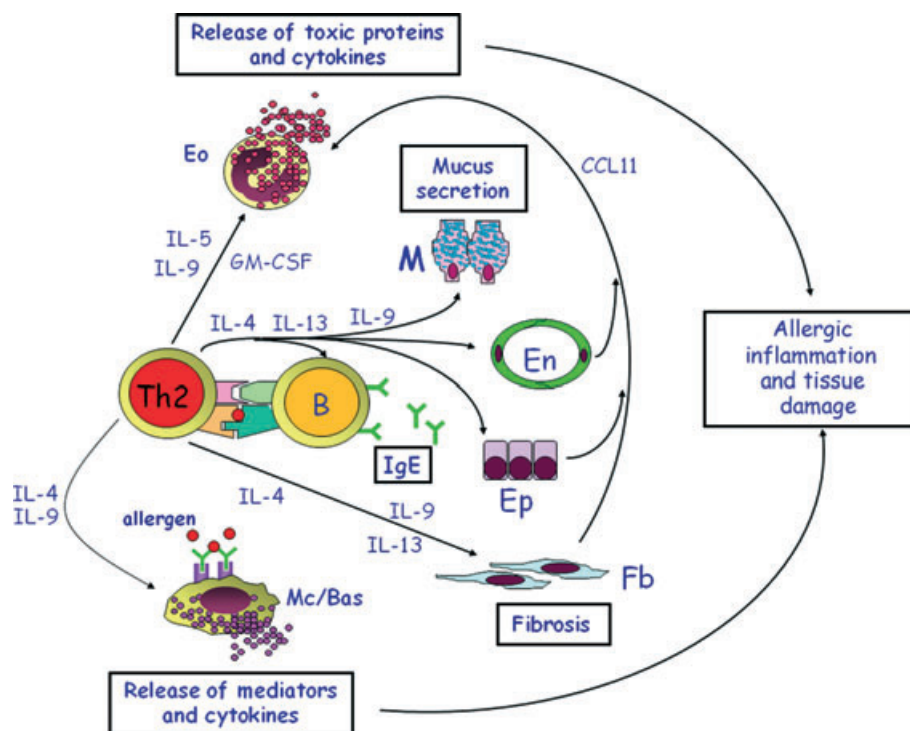


Figure 3 Role of Th2 lymphocytes in asthma pathogenesis. Cytokines and chemokines produced by Th2 cells, including GM-colony stimulatory factors, IL-4, IL-5, IL-9, IL-13, and those produced by other cell types in response to Th2 cytokines or as a reaction to Th2-related tissue damage (CCL11) account for most pathophysiological aspects of allergic disorders, such as production of IgE anti-

bodies; recruitment and activation of mast cells, basophils, and eosinophils granulocytes; mucus hypersecretion; subepithelial fibrosis; and tissue remodeling. B, B lymphocyte; Eo, eosinophil; Mc/Bas, mast cell/basophil; Ep, epithelial cell; M, mucus cell; En, endothelial cell; Fb, fibroblast. This figure has been adapted from Romagnani (89).

Identification of Th17/Th2 lymphocytes and their possible role in asthma pathogenesis

Recently we described a novel subset of human circulating memory CD4 T cells that produce both IL-17A and IL-4 (83). This previously unknown population of Th17/Th2 lymphocytes was more represented in the circulation of patients with allergic asthma than in healthy donors, suggesting a possible role in the pathogenesis of the disease. The existence of human Th17 cells able to produce IL-4, IL-5, IL-9, and IL-13 in addition to IL-17A, IL-8, and IL-22, was initially shown on T-cell clones generated from circulating CD4⁺ CD161⁺ CCR6⁺ T cells and then confirmed on freshly derived circulating CD4 T cells of healthy donors and asthmatics. The proportion of Th17/Th2 cells was extremely low in healthy subjects, whereas their numbers appeared to be significantly higher in the circulation of patients with chronic severe asthma. The proportion of Th17/Th2 cells was found to be significantly higher in Der p 1-, than in polyclonal-expanded, peripheral blood mononuclear cells (PBMC), derived from asthmatic Der p 1 allergic donors, suggesting that Th17/Th2 cells present in the circulation of atopic patients with asthma are specific for the sensitizing allergen. The observation that Th17 cells expressed the IL-4R, and were susceptible to the activity of IL-4, which induced STAT6 phosphorylation, whereas Th2 cells did not express IL-1RI and IL-23R, and did not show STAT3 or STAT4 phosphorylation in response to IL-23, led us to

hypothesize that an IL-4-rich microenvironment could favor the switch of allergen-specific Th17 lymphocytes toward the Th17/Th2 phenotype (83). Allergen-specific classic Th2 cells that respond to allergen in the lung could also modulate, thank to their ability to produce IL-4, Th17 lymphocytes specific for invading pathogens, toward the Th17/Th2 phenotype. This hypothesis is in keeping with our previous demonstration of the high plasticity of human Th17 cells (84). Another possibility is that allergen-specific Th2 lymphocytes become IL-17 producers in response to inflammatory stimuli. In fact it has been recently reported in mice that the proinflammatory cytokines IL-1 β , IL-6, and IL-21 could directly induce the up-regulation of *IRF4* and *ROR γ t* gene expression and the production of IL-17 in classical Th2 memory cells *in vitro*, suggesting a substantial phenotypic plasticity in response to inflammatory cues (85).

In any case, the demonstration of the existence *in vitro* and *ex-vivo* of CD4 cells able to produce both Th17-related and Th2-related cytokines, together with their increase in the circulation of patients with asthma, raises the important question of the pathophysiological role of this novel subset in allergic asthma. The presence of these cells at the level of bronchoalveolar lavage or bronchial biopsies of patients with asthma has never been reported in humans. However, the existence of the same subset of memory/effector T lymphocytes that coexpress the transcription factors GATA3 and ROR γ t and coproduce Th17 and Th2 cytokines has recently

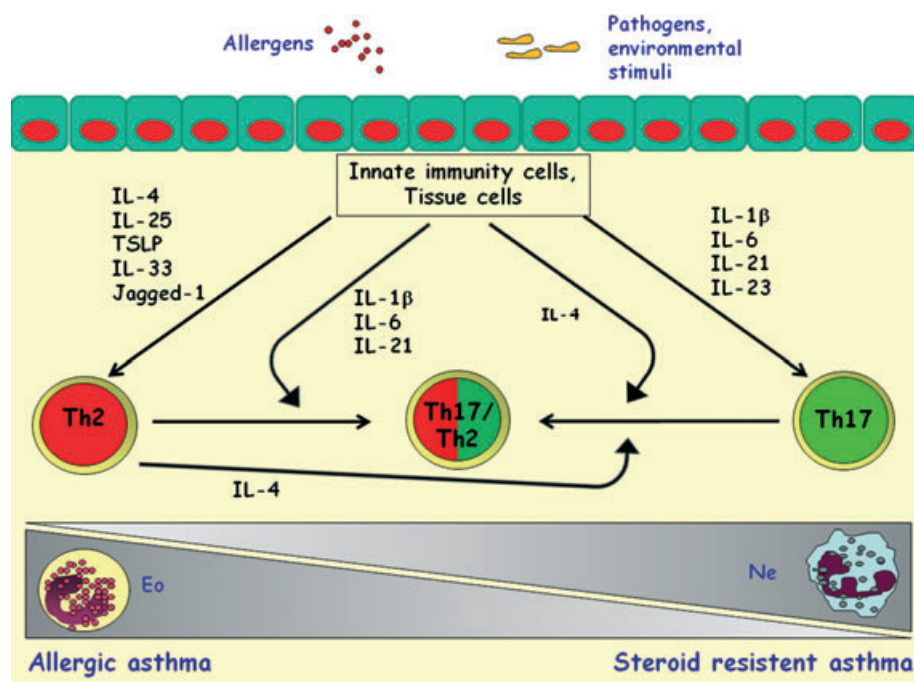


Figure 4 Th2 and Th17 lymphocytes support distinct phenotypes in asthma. Th2 and Th17 lymphocytes are the main actors of eosinophilic and neutrophilic forms of asthma, respectively. IL-4, IL-25, IL-33, TSLP, and Jagged-1 are important mediators of Th2-cell differentiation, while IL-1 β , IL-6, IL-21, and IL-23 are involved in the development of Th17 cells. The Th17/Th2 lymphocytes may origi-

nate from the activity of IL-4 produced by allergen-specific T lymphocytes, but probably also by innate immunity cells, on the Th17 cells, and/or from the activity of IL-1 β , IL-6, and IL-21 produced by innate immunity and tissue cells on the Th2 lymphocytes. Eo, eosinophil; Ne, neutrophil.

been reported in mice (85). In particular, in a mouse model of induced asthma, transfer of allergen-specific, IL-17-producing Th2 cells resulted in profound goblet hyperplasia as well as elevated mucin production after antigen sensitization with heterogeneous leukocytes infiltrating the airways, including neutrophils, eosinophils, macrophage, and lymphocytes. In contrast, mice transferred with conventional Th2 or Th17 cells exhibited fewer airway infiltrations of eosinophils or neutrophils, respectively, and limited pathophysiological features (85). Recently, Lajoie et al. (86), by using a well-characterized mouse model of differences in susceptibility to severe asthma, showed that the strain of mice that develops severe AHR (A/J mice) produced both IL-17A and Th2 cytokines, whereas the strain that manifests less-severe AHR (C3H/HeJ mice) produced only Th2 cytokines and little to no IL-17A. Blockade of IL-17A in susceptible (A/J) mice decreased the severity AHR, whereas reconstitution of IL-17A in C3H/HeJ mice exacerbated AHR. Consistent with the widely acknowledged importance of Th2 cytokines in driving the development of allergen-induced AHR, IL-17A alone was unable to induce AHR. However, the simultaneous production of IL-17A and Th2 cytokines exacerbated Th2-driven pathology, which led to more severe disease. Mechanistically, the production of IL-17A and development of robust AHR was reciprocally regulated by anaphylatoxins, with signaling by complement factor C5a limiting the frequency of Th17 cells and AHR, and signaling by complement factor C3a enhancing Th17 responses and AHR. The opposing actions of C3a and C5a on IL-17A were mediated via reciprocal regulation of DC production of IL-23 (86).

Possible future applications

In the last years, it has become evident that asthma is an extremely heterogeneous disease with many clinical pheno-

types characterized by distinct pathogenic mechanism, whose knowledge will allow approaching asthma therapy from a new perspective. These phenotypes, including allergic asthma, severe steroid-resistant asthma, asthma associated with obesity, induced by exercise, exposure to air pollutants or to aspirin, are characterized by different cellular networks supporting the chronic inflammation. The role of Th2 lymphocytes and eosinophils infiltrating the lung in the pathogenesis of the allergic form of the disease is supported by several evidences (3), and more recently, it has been suggested that Th17 lymphocytes and neutrophils could be involved in nonatopic (70) and in steroid-resistant (87) asthma. The definition of different asthma phenotypes is crucial for redirecting the therapeutic approach, which in future will be hopefully personalized for each patient. It is well known that steroids have little effect in neutrophilic asthma, whereas they work fine in eosinophilic asthma, in which also anti-interleukin-5 antibodies seem to have some benefit (88). Although these pathways can develop independently, they can also coexist in the same patient, with the consequence that the therapy for asthma could be planned in future also in a phase-specific manner (Fig. 4). In this view, it results particularly important not only the understanding of the factors that regulate the balance of different T-helper subsets, but also their temporal sequences and potential interactions in the induction of inflammation.

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Conflict of interest

The authors declare that they have no conflict of interest.

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