Regulation in chronic obstructive pulmonary
disease: the role of regulatory T-cells and
Th17 cells

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ABSTRACT

COPD (chronic obstructive pulmonary disease) is an inflammatory disorder of the airways, which is associated with irreversible airway obstruction. The pathological hallmarks of COPD are destruction of the lung parenchyma (pulmonary emphysema), inflammation of the central airways (chronic bronchitis) and inflammation of the peripheral airways (respiratory bronchiolitis). Tobacco smoking is established as the main aetiological factor for COPD. A maladaptive modulation of inflammatory responses to inhalation of noxious particles and gases is generally accepted as being a key central pathogenic process; however, the precise regulatory mechanisms of the disease are poorly understood. Two cell types are known to be important in immune regulation, namely regulatory T-cells and the newly identified Th17 (T-helper 17) cells. Both types of cells are subsets of CD4 T-lymphocytes and modulate the immune response through secretion of cytokines, for example IL-10 and IL-17 respectively. The present review will begin by describing the current understanding of inflammatory cell involvement in the disease process, and then focus on the possible role of subsets of regulatory and helper T-cells in COPD.

DEFINING THE DISEASE

COPD (chronic obstructive pulmonary disease) is a major cause of ill-health and mortality, predicted to become the third commonest cause of death worldwide by 2030 [1]. It is predominantly caused by smoking and is characterized by poorly reversible airflow limitation. The cardinal pathological features are small airways disease (obstructive bronchiolitis), mucus hypersecretion and destruction of the lung parenchyma (emphysema), which lead to the main symptoms of chronic cough, sputum production and disabling breathlessness. The underlying pathology is caused by an “abnormal inflammatory response of the lung to noxious particles or gases” [2] (http://www.goldcopd.org/Guidelineitem.asp?l1=2&l2=1&intId=2003) with the key inflammatory cells and mediators of disease being neutrophils, macrophages and T-lymphocytes, along with the mediators they secrete, such as elastase, MMPs (matrix metalloproteases) and cytokines (Figure 1).

Key words: chronic obstructive pulmonary disease (COPD), inflammation, regulatory T-cell, regulatory type 1 cell, T-helper 17 cell.

Abbreviations: APC, antigen-presenting cell; BALF, bronchoalveolar lavage fluid; CCR, CC chemokine receptor; COPD, chronic obstructive pulmonary disease; CTLA, cytotoxic T-lymphocyte antigen; CXCL, CXC chemokine ligand; FOXP3, forkhead box P3; IFN-γ, interferon-γ; IL, interleukin; IL-17R, IL-17 receptor; IL-23R, IL-23 receptor; MMP, matrix metalloprotease; NK cell, natural killer cell; NKT cell, NK T-cell; iNKT cell, invariant NKT cell; RORγt/RORC2, retinoic orphan receptor γt; TCR, T-cell receptor; TGF-β, transforming growth factor-β; TGF-βR, TGF-β receptor; Th, T-helper; TNF, tumour necrosis factor; GITR, glucocorticoid-inducible TNF receptor; Tr1 cell, regulatory type 1 cell; Treg-cell, regulatory T-cell; iTreg-cell, adaptive/induced Treg-cell; nTreg-cell, natural Treg-cell.

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In the present review we will briefly summarize the evidence regarding inflammatory cell involvement in the disease process. We will then focus on the possible role of subsets of T\textsubscript{reg}-cells (regulatory T-cells) and Th (T-helper) cells in COPD. These include nT\textsubscript{reg}-cells (natural T\textsubscript{reg}-cells), iT\textsubscript{reg}-cells (adaptive/induced T\textsubscript{reg}-cells), Tr1 cells (regulatory type 1 cells) and Th17 cells (T-helper 17 cells). Their function and phenotype will be described and we will hypothesize on their potential role in COPD.

**Inflammatory Cell Involvement**

Evidence for involvement of neutrophils in COPD includes the finding of higher numbers in sputum, BALF (bronchoalveolar lavage fluid) and airway smooth muscle [3] of patients with COPD. The finding that neutrophil numbers in bronchial biopsies and induced sputum correlate with disease severity [4] and rate of decline in lung function [5] adds to the evidence for their potential importance. Neutrophils are attractive candidates for effector cells since they release a number of mediators that cause damage to both the airway and lung parenchyma. Historically, neutrophil elastase was seen as one of the chief candidates for causing lung damage, but other mediators, such as cathepsins and MMPs, are also important. Furthermore, neutrophils may stimulate mucous secretion by up-regulating mucin expression through the production of neutrophil elastase and reactive oxygen species [6].

Macrophages also have the potential to contribute to the pathology of the disease through the release of MMPs, predominantly MMP9, and other tissue-destructive enzymes, such as collag enase 1 and 2 and gelat inases A and B [7]. Macrophage numbers are increased in the sputum and BALF of smokers with COPD and bronchial biopsy studies, and examination of resected lung tissue shows there to be increased numbers in the airway wall and alveoli [8,9]. Macrophages are thought to be the major source of MMPs in the airway, and increased levels of MMPs have been found in both the sputum and BALF of patients with COPD compared with healthy non-smokers [10]. MMPs are able to degrade nearly all of the components of the extracellular matrix and could clearly cause the characteristic lung destruction and airway damage.

Work on T-lymphocytes has mainly focused on the role of the CD8 cell. Early studies showed increased numbers of these cells in the bronchial biopsies of smokers with COPD [11] compared with healthy smokers, and increased numbers of CD8 T-cells have subsequently been shown in peripheral airways [12], lung parenchyma and even paratracheal lymph nodes [13]. Knockout studies have suggested a critical role for these cells since CD8\textsuperscript{−} mice fail to develop emphysema when exposed to cigarette smoke [14]. The contribution these
cells make to the inflammatory pathway is less clear. One possible mechanism is through perforin- and granzyme-mediated cytotoxicity. Increased levels of perforin have been shown in sputum from patients with COPD and increase in T-cell expression of these mediators has been demonstrated in the BALF [15]. Other possible roles of CD8 cells include release of cytokines, such as IL- (interleukin)-13 and IL-17 that can up-regulate MMP expression [16,17] and release of the pro-inflammatory cytokine IL-18 [18]. In a mouse model of COPD, CD8 T-cells have been shown to contribute to macrophage recruitment [19].

There are conflicting reports on the possible role of two other types of killer cells, namely NK cells (natural killer cells) and NKT cells (NK T-cells), in COPD [20–22]. We have shown increases in perforin and granzyme expression and increased cytotoxicity of NK (CD56\(^+\)CD3\(^-\)) and NKT-like (CD56\(^+\)CD3\(^+\)) cells from the induced sputum of patients with COPD [23], and have shown previously reciprocal changes in the blood [24], suggesting the migration of more cytotoxic cells into the lung. This could contribute to the previously described increases in perforin and granzyme levels in the sputum of COPD patients but, as these killer cells can also stain for CD8, some of the activity attributed previously to CD8 T-cells in COPD could be due to these other killer cells.

Increases in CD4 cells have been found in the peripheral airways of smokers with COPD [25], and lung CD4 cells increase significantly after 30 years of smoking [26]. Again their role is unclear, although their main function is cytokine secretion. CD4\(^+\) Th2 cells from the central airways of smokers with chronic bronchitis stain for IL-4 and IL-13 [27], and these CD4\(^+\) Th2 cells are able to stimulate mucus production through IL-13 [28]. Alternatively IFN-\(\gamma\) (interferon-\(\gamma\)) produced by CD4\(^+\) Th1 cells could contribute to the inflammatory process through activation of macrophages and, indeed, IFN-\(\gamma\)-overexpressing transgenic mice develop emphysema [29]. It is now known that CD4 cells are functionally more diverse than implied by the traditional Th1/Th2 phenotypes, and this is controlled both thymically and peripherally.

**T-CELL DEVELOPMENT**

T-lymphocyte progenitors originate in the bone marrow, but their development occurs in the thymus. These developing T-cells receive a signal (most probably from stromal cells in the thymus) that is transduced through Notch1, which is thought to regulate the T-cell lineage [30]. These thymocytes develop into one of three main T-cell types in the thymus, namely naïve CD8 T-cells, naïve CD4 T-cells and NKT-cells, through interaction with dedicated APCs (antigen-presenting cells). More recently, it has been shown that thymocytes can also develop into nTreg\(_{\text{cells}}\) in the thymus as a result of a T-cell intrinsic control mechanism, which does not depend on APCs [31] (Figure 2).

NKT cells and nTreg\(_{\text{cells}}\) leave the thymus primed ready to carry out their effector functions (i.e. cytokine secretion or mediator release), whereas naïve T-cells (CD4 or CD8) require further activation before they become effector cells. These naïve T-cells are primed by pathogen-activated APCs through their TCR (T-cell receptor). Three kinds of signals are required for naïve T-cell activation by APCs. The first signal is the TCR-MHC interaction and relies on the appropriate peptide being displayed by the MHC molecule on the surface of the dendritic cell. In the case of CD8 T-cells, MHC class I is required, whereas for CD4 T-cells MHC class II is necessary. Signal two is a costimulatory signal, exemplified by the interaction of CD80 with CD86 on the surface of the APCs, whose net effect is increased survival and proliferation. Signal three is commonly thought to determine the type of T-cell generated, particularly in the case of CD4 T-cells (Figure 2). The cytokines secreted by APCs or the environmental milieu at the time of interaction is generally, but not exclusively, thought to direct T-cell functional differentiation. In the presence of IL-12, naïve CD4 T-cells become Th1 cells, whose main function is viral clearance [32]. Th2 cells are generated when IL-4 is predominant and these cells are mainly involved in allergic responses [33]. In an IL-2 rich environment, iTreg\(_{\text{cells}}\) or Tr1 cells are generated [34] and, in the presence of IL-6 and TGF-\(\beta\)1 (transforming growth factor-\(\beta\)1), Th17 cells develop [35] (Figure 2).

**REGULATORY T-CELLS**

**Phenotype**

The theory of T-cell-mediated regulation first emerged with the concept of suppressor T-cells [36] and has now re-emerged with the identification of Treg\(_{\text{cells}}\). These cells are thought to play a major role in regulating the immune system and there are currently three known cell subsets: nTreg\(_{\text{cells}}\), iTreg\(_{\text{cells}}\) and Tr1 cells (Figure 3).

nTreg\(_{\text{cells}}\) leave the thymus as effector cells, whereas the iTreg\(_{\text{cells}}\) and Tr1 cells mature in the periphery. nTreg\(_{\text{cells}}\) are the most abundant subset and they represent 5–10% of the CD4\(^+\) T-lymphocyte population [37]. They constitutively express CD25 (IL-2Rα (IL-2 receptor α) chain), GITR (glucocorticoid-inducible TNF (tumour necrosis factor) receptor), CTLA4 (cytotoxic T-lymphocyte antigen 4), a T-cell-inhibitory receptor [38], and FOXP3 (forkhead box P3). FOXP3 is known as a master regulator of Treg\(_{\text{cells}}\) and is one of the most commonly used markers to identify nTreg\(_{\text{cells}}\). FOXP3 is a forkhead winged helix transcription factor, but it has
The current hypothesis of CD4 T-lymphocyte differentiation

The maturation of iTreg-cells and Tr1 cells occurs after leaving the thymus. There is potential to switch from the Treg-cell response to a Th17 response under an appropriate microenvironment, as well as Th17 to switch to Th1 or Th2.

been shown not always to be necessary for suppressive function [39] as it is not expressed by all Treg-cell subsets. nTreg-cells express a variety of other markers, including CD39, LAG3 (lymphocyte-activation gene 3), OX40 (CD134), integrins and chemokines. The cells also express chemokine receptors, including CCR (CC chemokine receptor) 6 and CCR7, which give them the ability to migrate to sites of inflammation where they can carry out their regulatory function [40] (Figure 3A).

iTreg-cells display similar markers to nTreg-cells once they have matured in the periphery (i.e. FOXP3, CD25, CTLA4 and GITR). Their development relies on the presence of TGF-β1 converting them from CD25− into a constitutive CD25+ phenotype [41]. TGF-β1 also provides a positive-feedback loop to enable population expansion (Figure 3B).

Tr1 cells are the most poorly defined subset. They too mature in the periphery upon antigen activation via the TCR [42]. The main difference between the two other types of regulatory cells is that Tr1 cells are thought to be CD25-negative and FOXP3-negative or low [43]. Tr1 express high levels of anti-inflammatory cytokines such as IL-10; this cell type is also known to secrete IL-5 and IFN-γ (Figure 3C).

Other T-cells with regulatory function

A number of other cells have been described with regulatory function. For example, a subset of CD8+ T-cells can be induced to become regulatory CD8 T-cells by the co-stimulatory molecule 4-1BB [44,45]. A further subset of CD8 T-cells, namely CD8+CD28null T-cells, may have a regulatory role although, under certain conditions, they may also be pro-inflammatory [46]; however, these cells have limited TCR diversity [47]. Other regulatory CD4 cells include Th3 cells, which are TGF-β-producing cells induced in oral tolerance [48].

T-cells that recognize antigen presented by the MHC class 1b molecule CD1d are known as iNKT (invariant NKT) cells [49] and these are known to have a regulatory role [50].

Control of differentiation

As already discussed, the cytokine microenvironment at the time of initial T-cell activation guides the functional differentiation of the T-cell response generated. This may thus be influenced by the level of inflammation and cytokine milieu observed in smokers with COPD.

TGF-β1 and IL-2 are the two crucial cytokines involved in the differentiation of naive T-cells into Treg-cells. TGF-β1 together with IL-2 are also needed for Treg-cell expansion. IL-2-deficient mice have a reduced number of nTreg-cells, indicating the importance in Treg-cell proliferation [52]. Increased levels of TGF-β1 in serum in patients with COPD compared with healthy controls has been found [53]. In addition, IL-2 levels are higher in patients with stable COPD [54] compared with those who exhibit a rapid decline in lung function. The findings of higher levels of both TGF-β1 and IL-2 suggest...
that an environment exists in COPD that encourages differentiation into T_{reg} cells and this may be particularly pronounced in those patients with stable disease.

**T_{reg} cell function, mediators and possible role in COPD**

The role of T_{reg} cells is to provide essential protection to the body from an overactivated immune response. T_{reg} cells are important both for the production and induction of anti-inflammatory cytokines in chronic inflammation, such as in COPD. They achieve this by acting via cytokines or by recruiting other cell types, such as neutrophils and APCs (namely macrophages and dendritic cells). The functions of T_{reg} cells are varied in their nature. nT_{reg} cells are essential in maintaining self-tolerance [55], and iT_{reg} cells and Tr1 cells can be stimulated when the immune system is responding to pathogens [56]. The differentiation of T_{reg} cells is due to the presence of mediators, co-stimulatory molecules and the type of antigen encountered. nT_{reg} cells encounter autoantigens; hence one of their main roles is to control self-reacting T-cells. They do this by contact-dependent mechanisms or via bystander suppression, whereby the cytokines they release have effects on other cell types. Exogenous antigens activate iT_{reg} cells and Tr1 cells. Very low levels of antigens can stimulate iT_{reg} cells, whereas Tr1 cells respond to a high antigenic load, such as in severe inflammation where the exposure can be chronic.

nT_{reg} cells produce the novel anti-inflammatory cytokine IL-35, which inhibits T-cell proliferation. IL-35 is a heterodimer that is part of the IL-12 cytokine family along with IL-23 and IL-27 [57]. In mice, IL-35 is involved in the suppression of Th17 cells [58], but the same function has not been demonstrated in humans [59]. To date, IL-35 has been shown to be secreted by T_{reg} cells but not other effector T-cells (i.e. Th cells or Tc cells (cytotoxic T-cells)). The receptor for IL-35 is not known, but it is reasonable to assume that it may share
receptors of other siblings in the IL-12 family and these are expressed on macrophages, dendritic cells, monocytes and B-cells [60]. One could therefore hypothesize that IL-35 could regulate the functions of cells known to be important in COPD, specifically macrophages.

iTreg-cells are known for their production of TGF-β so, in this context, play an anti-inflammatory role. TGF-β production by iTreg-cells therefore could have a regulatory role in the pathogenesis of COPD due to expression of TGF-β receptors on macrophages, epithelial cells and fibroblasts [61], all important cell types in the pathophysiology of COPD.

The main function of Tr1 cells is to secrete the anti-inflammatory cytokine IL-10 [62], which is also secreted by monocytes. IL-10 has the ability to suppress Th1 cytokines such as IFN-γ and TNF-α and has anti-inflammatory effects on neutrophils by inhibiting IL-8 and MIP-1 (macrophage inflammatory protein-1). In COPD, neutrophilia is a key feature, and this may suggest that IL-10 production by Tr1 cells may not be sufficient to prevent neutrophilic infiltration in these individuals. It would therefore be of interest to further examine IL-10 production by these cells in COPD.

In the absence of IL-10, IL-23 levels can be elevated leading to the activity of the Th17 lineage [63]. IL-27 is another novel cytokine which may play a role in tissue inflammation, as IL-27 promotes IL-10-secreting Tr1 cells and inhibits the Th17 pathway [64].

From our understating of iTreg-cell function, one could hypothesize that iTreg-cell numbers would be reduced in COPD, leading to the inflammation observed in disease. Interestingly, increased levels of iTreg-cells have been found in COPD patients with acute exacerbations [65] and in the lungs of patients with emphysema [66]. Previously, the number of CD4+CD25+ T-cells in BALF was directly correlated with the number of pack years in COPD patients [67], but this has not been linked directly to disease. Other groups have concluded that there is a direct positive correlation between number of FOXP3+ cells stained in the large airways and the number of pack years in COPD patients [68]. CD4+CD25+ cells have been reported to be significantly increased in BALF compared with peripheral blood across healthy volunteers, smokers and COPD patients [69]. More recently, a study on lung parenchyma of moderate COPD patients concluded that there is an increased number of CD4+FOXP3+ in lymphocyte follicles [70]. However, an increase in iTreg cells appears to be related to smoking rather than disease [71].

Overall, the evidence indicates that there is an increase in iTreg-cells in COPD patients (Table 1), but these cells may not be effective in regulating inflammation in the lung; the outcome will clearly depend on the balance between pro- and anti-inflammatory influences. Regulatory functions may be insufficient due to a dysfunction of the regulatory cells. Consistent with this idea, smokers who do not develop COPD also have elevated iTreg-cell numbers in the lung and these cells may be more effective at regulating the inflammatory response to cigarette smoke (see Figure 5).

Macrophages and their interactions with iTreg-cells are complex. In normal homoeostatic conditions, macrophages and other APCs, such as dendritic cells, are kept under control by iTreg-cells via apoptosis or by a direct inhibition of their activation and therefore function [72]. However, in chronic disease, APCs may escape regulation and go on to produce pro-inflammatory cytokines which can induce a Th17 response. This may be the case in COPD, as increased numbers of macrophages play an important role in the disease pathology; they are especially involved in lung parenchyma destruction and structural changes in the small airways.

Table 1 Expression data of CD4+ Treg-cells and Th17 cells in COPD

<table>
<thead>
<tr>
<th>Cell type</th>
<th>Expression</th>
<th>Compartment</th>
<th>Reference</th>
</tr>
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<tbody>
<tr>
<td>iTreg-cell</td>
<td>↑nTreg-cells in exacerbations of COPD correlated with severity</td>
<td>Peripheral blood</td>
<td>[65]</td>
</tr>
<tr>
<td></td>
<td>↑nTreg-cells in smoking-induced emphysema</td>
<td>Lung tissue</td>
<td>[66]</td>
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<td></td>
<td>Correlation between CD4+CD25+ T-cells and pack years</td>
<td>BALF</td>
<td>[67]</td>
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<tr>
<td></td>
<td>Correlation between Foxp3+ cells and pack years of COPD patients</td>
<td>Large airways (surgical specimens)</td>
<td>[68]</td>
</tr>
<tr>
<td></td>
<td>↑CD4+ Foxp3+ cells in moderate COPD compared with smokers with normal lung function and healthy controls</td>
<td>Lymphocyte follicles</td>
<td>[70]</td>
</tr>
<tr>
<td>Th17 cell</td>
<td>↑Levels of IL-17 in murine models exposed to cigarette smoke</td>
<td>Murine lung and BALF</td>
<td>[104,105]</td>
</tr>
<tr>
<td></td>
<td>↓Smad 7 in bronchial biopsies of COPD patients compared with healthy non-smokers</td>
<td>Bronchial biopsies</td>
<td>[108]</td>
</tr>
<tr>
<td></td>
<td>No change in RORC2 cells in smokers with and without COPD</td>
<td>Bronchial biopsies</td>
<td>[103]</td>
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Th17 CELLS

Phenotype

The IL-17 cytokine was identified more than 15 years ago and was originally named CTLA8 [73]. In 1995, it was first reported that CD4+ T-cells were capable
of producing IL-17 independently of the Th1 and Th2 subset lineage [74] and, consequently, the Th17 family of cells emerged. The Th17 lineage is defined as CD4+ T-lymphocytes that predominantly secrete the cytokine IL-17 [75]. RORγt (retinoic orphan receptor γt) is the transcription factor, with its human homologue RORC2, that is required for the development of Th17 cells [76]. Th17 cells characteristically express TGF-βR1 (TGF-β receptor 1), IL-6R (IL-6 receptor) and IL-23R (IL-23 receptor) on their cell surface. There are also a variety of chemokine receptors expressed on the cell surface, such as CCR4 and CCR6, which are involved in homing to inflamed tissue (Figure 4).

**Figure 4** Th17 cell phenotype and receptors involved in the differentiation and stabilization of the lineage
A variety of cytokines are associated with the Th17 cell, including IL-17A, IL-17F, IL-21, IL-22 and IL-6 and, to a lesser extent, IFN-γ and TNF-α.

**Control of differentiation**

The cytokine microenvironment again influences the differentiation of Th17 cells. The Th17 lineage depends on the presence of TGF-β1 and IL-6 at the optimum concentrations [77]. TGF-β1, IL-23, IL-1β and IL-6 all contribute to human Th17 differentiation [78]. TGF-β1 in this context has a dual function: in the presence of IL-6 it is capable of inducing Th17 cells, but when IL-6 is absent it induces Treg-cell development. Thus TGF-β1 has the ability to stimulate the most appropriate response, whether it is the expansion of regulatory cells or up-regulation of Th17 cells [79].

TGF-β1 is traditionally thought as an anti-inflammatory mediator; however, recently it has been implicated as being essential for Th17 differentiation. TGF-β1 is released from Treg-cells, dendritic cells and macrophages. Th1 and Th2 differentiation is inhibited by TGF-β1, which in turn encourages Th17 differentiation. TGF-β causes alveolar macrophages to release pro-inflammatory cytokines and MMPs upon exposure to cigarette smoke in emphysema [80].

IL-6 can act as a regulatory cytokine, and it has the ability to shift the balance from an innate to an adaptive immune response [81]. This ability may play a pivotal role in the potential switching of Treg-cells (which, as discussed, influences neutrophil and macrophage recruitment and activation) into a Th17 response (which primarily influences T-cells) in the presence of chronic infection. Effective anti-IL-6 treatments have been used in other inflammatory diseases, namely rheumatoid arthritis and Crohn’s disease [82]. IL-6 also induces IL-21, which is required for Th17 differentiation. In the absence of IL-6, TGF-β is switched from the Th17 route to the Treg-cell pathway [79]. New evidence suggests that Treg-cells can co-express FOXP3 and IL-17 [83], suggesting plasticity between lineages.

As described previously, TGF-β1 is elevated in serum from COPD patients [53], which could lead to increased numbers of Th17 cells. Serum and sputum levels of IL-6 are increased during periods of exacerbations in COPD [84], again potentially leading to increased numbers of Th17 cells. Furthermore, increased staining of IL-23, another promoter of Th17 differentiation, has been observed in patients with COPD [103].

**Th17 function, mediators and possible role in COPD**

The Th17 response induces pro-inflammatory cytokines and chemokines, such as CXCL (CXC chemokine ligand) 8 and CXCL10 from a variety of cell types (such as macrophages and epithelial cells), thereby recruiting neutrophils to inflamed tissues [85]. Cigarette smoke can lead to a direct up-regulation of many chemokines associated with Th17 cells, such as CXCL8 and CCL20, which are involved in promoting neutrophil inflammation in the lung [86]. Other chemokines, such as CCL5 and CXCL7, that are also recognized as Th17 markers are increased in the bronchial mucosa of patients with COPD [87], and so may be involved in the attraction of leucocytes and neutrophils to inflammation in the lung.

The key cytokine secreted by Th17 cells is IL-17 itself. The IL-17 family consists of six cytokine members IL-17A–IL-17F [88], and five receptors IL-17RA–IL-17RE [89]. IL-17A and IL-17F are 50% homologous and share similar functions. IL-17 is secreted from a variety of cells, including NKT cells, macrophages, dendritic cells, B-cells and γδ T-cells [90]. IL-17RA is the largest member of the IL-17R family and at least four ligands are mediated through this subunit [91]. The expression of IL-17RA is up-regulated by cytokines such as IL-15 (produced by a range of non-T-cells, including macrophages [92]) and IL-21 [93,94] on CD8+ T-cells, and so may play an important role in COPD.

IL-21 and IL-22 are two further cytokines secreted from Th17 cells. IL-21 is necessary to provide the
positive-feedback loop for successful amplification of Th17 cells [95], and is required for the survival of a range of immune cells such as lymphocytes, NK cells and B-cells.

IL-22 is produced not only by Th17 cells, but also by other T-cell subsets, NK cells and activated dendritic cells. The IL-22R (IL-22 receptor) is expressed on epithelial cells and fibroblasts within tissues. IL-22 has been detected in a variety of organs such as intestines and lung [96]. The up-regulation of IL-22 has been linked to disease severity in other inflammatory disorders, such as psoriasis [97,98], rheumatoid arthritis [98] and irritable bowel disorder [99]. Alternatively IL-22 can provide protection against certain infections and induce the expression of β defensins, which are important in the protection of mucosal barriers against microbes in the lung [100]. Further work examining the levels of IL-22 in patients with COPD could show whether changes in IL-22 may contribute to their recurrent infections.

IL-23 plays a key role in the maintaining and expanding the Th17 cell lineage over time to release IL-17 and IL-22 [35]. IL-23 is secreted from APCs such as dendritic cells and is part of the IL-12 cytokine family. TGF-β1 induces the IL-23R (IL-23 receptor) on Th17 cells [77]. IL-23R knockout mice develop less severe symptoms in experimental autoimmune encephalitis and inflammatory bowel disease models, implying a role in chronic inflammation [101]. IL-23 is released in response to inflammatory signals and therefore is continually expressed in chronic inflammation. IL-23 has also been shown to enhance eosinophilic inflammation in a murine asthma model [102]. IL-22 and IL-23 have been detected in patients with stable COPD by immunohistology staining of the bronchial epithelium and submucosa. Furthermore, they demonstrated that the increase in IL-22-positive cells could be significantly correlated with the number of CD4+ and CD8+ cells in patients with COPD and smokers [103]. Analysis of other samples, such as peripheral blood and BALF, would be useful in determining the role of these cytokines in COPD.

There has been little investigation of the role of IL-17 in the lungs, especially in COPD. Current evidence using a murine model has shown increased levels of IL-17 in lung homogenate exposed to cigarette smoke [104,105]. Murine lung epithelial cells have also shown that overexpression of IL-17A induces COPD-like lung inflammation [106]. Increased levels of IL-17 mRNA have been detected in the sputum of asthmatics [107], but these studies have not been demonstrated in patients with COPD. However, studies in COPD have shown a down-regulation of Smad7, a signalling molecule involved in the Th17 pathway in bronchial biopsies [108]. Most recently, a study concluded that there were increased levels of Th17-related cytokines in stable COPD patients but this may be due to smoking [103]. An increased number of IL-17A-, IL-22- and IL-23-positive bronchial epithelial cells were found in stable COPD patients compared with healthy controls; however, the number of RORC2-positive cells was not significantly different between smokers and COPD, implying that any differences may be due to smoking alone [103]. Human Th17 cells differ from other subsets in their potency to induce pro-inflammatory cytokines in bronchial epithelial cells, suggesting a role in inflammatory airway diseases. Th17 cells produce more inflammatory cytokines, such as IL-6 and IL-1β, compared with Treg-cells and other Th cells [109].

Interestingly, increased levels of IL-6 and TNF-α are found in sputum and BALF and have been associated with disease severity in patients with COPD [110]. TNF-α promotes CXCL8 expression from airway epithelial cells. Increases in serum TNF-α has also been linked to exacerbations in COPD patients [111]. In addition TNF-α production by mast cells is increased due to IL-17A, leading to neutrophil infiltration in the airways [112]. IL-17 is capable of increasing mucin production from airway epithelial cells [17]; hypermucus production is one characteristic of COPD. IL-17 can also induce GM-CSF (granulocyte/macrophage colony-stimulating factor) from airway epithelial cells [113], which plays an important role in recruiting APCs to control infection. IL-17A from Th17 cells has also been implicated in neutrophilic inflammation in the airways. An increase in neutrophils and IL-17 have been linked in COPD [114]. Mice treated with an anti-IL-17A antibody had a reduced recruitment of airway neutrophilia in BALF samples [115].

Overall, the evidence regarding Th17 cells in COPD is mixed, with some reporting a reduction in these cells [108], whereas others reporting increases in Th17 cytokine staining [103] (Table 1). This leaves the function of these cells in COPD open to debate.

CONCLUSIONS

In conclusion, Treg-cells are known to maintain peripheral tolerance, regulate APCs and provide an immunosuppressive role via anti-inflammatory cytokines. Conversely, Th17 cells produce pro-inflammatory cytokines so may play a role in chronic inflammatory disorders. In COPD, it is possible to hypothesize that the body’s regulatory system is not effective in preventing the lung damage caused by the effects of cigarette smoke and subsequent inflammation; therefore the immune system switches to a Th17 response which is pro-inflammatory (Figure 5). Much further work needs to be done, in particular, the function and numbers of these cells in the airways and greater clarification of their role in acute and stable disease and during exacerbations.
The type of immune response activated relies on the cytokine environment and the individual's immune system, leading to varied clinical outcomes. In some situations, there can be a switch of lineage from Treg cells to Th17. TGF-β and IL-6 are crucial in the transition between lineages.

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