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Biologic and pharmacologic therapies in clinical development for the inflammatory response in COPD

Jaymin B. Morjaria[1], Mario Malerba[2] and Riccardo Polosa[3]

[1] Department of Infection, Inflammation and Repair, University of Southampton, Southampton SO16 6YD, UK
[2] Department of Internal Medicine, University of Brescia, Spedali Civili 1, 25100 Brescia, Italy
[3] Direttore U.O.C. di Medicina Interna, Az Ospedaliero-Universitaria Vittorio, Emanuele - P>O>S Marta, Via G. Clementi 36, 95124 Catania, Italy

Chronic obstructive pulmonary disease (COPD) is a progressive and debilitating condition with declining lung function associated with airway inflammation, mucus hypersecretion and remodeling. Inflammatory cells contribute to the disease processes via the production of proteases, fibrotic or mitogenic growth factors, cytokines, chemokines and their receptors. Particularly newer agents that treat the underlying inflammation and remodeling. Here, we briefly review current understanding of the inflammatory mechanisms involved in the pathogenesis of COPD. This understanding has enabled the identification of several therapeutic targets that might have great potential for the development of novel anti-inflammatory and anti-remodeling biologic therapies with considerable clinical advantage for COPD. Some of these molecules have been assessed, and others are in the early stages of being assessed. This article gives an up-to-date summary of these novel therapies and their status of clinical development in targeting the various inflammatory pathways of COPD.

Introduction

Chronic obstructive pulmonary disease (COPD) is a progressive and debilitating disease that affects millions of people and is characterized by a persistent inflammatory response that cannot be reversed and generally leads to progressive decline in lung function, respiratory failure, cor pulmonale and death [1]. The persistent inflammatory response of the airways in COPD is typically associated with smoking [2], but environmental pollutants are also important causes in developing countries [3]. Studies have reported that smokers have a more rapid rate of decline in forced expiratory volume in 1 s (FEV_1) [4], and once inflammation is established in COPD, it persists even when smoking has stopped. This inflammation of the airways in COPD responds marginally to current anti-inflammatory drugs, including corticosteroids [5,6]. Thus – with the exception of smoking cessation, which is known to effectively slow lung function decline; long-term oxygen therapy, which seems to prolong survival; and bronchodilators, which might briefly reduce symptoms there is a compelling need for improved therapeutic strategies for patients with COPD, particularly therapies that specifically target the most relevant inflammatory pathways.

The inflammatory infiltrate in the airways or alveoli of patients with COPD is characterized by elevated numbers of neutrophils, monocytes, macrophages, $CD8^+$ and $CD4^+$ T cells, B cells, dendritic cells, natural killer cells and/or mast cells in the airway walls, alveolar compartments and vascular smooth muscle [7–9]. This chronic inflammation of the lung typically results in disease progression leading to mucus hypersecretion and structurally irreversible lung lesions. As our understanding of the pathology of COPD has increased, it has been established that the progressive pulmonary inflammation that is associated with COPD is likely to be a key target for novel therapeutics. Here, in addition to giving an overview of the inflammatory mechanisms involved in the pathogenesis of COPD, we specifically consider novel biologic and pharmacologic therapies that are currently in clinical development for COPD and discuss how these drugs might modulate pulmonary inflammation and remodeling in this disease.

Corresponding author:. Morjaria, J.B. (jbm@soton.ac.uk)

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Inflammatory cells involved in the pathogenesis of COPD

The innate defense system of the lung includes the mucociliary clearance apparatus, the epithelial barrier, and the coagulation and inflammatory cascades [8,10]. Smoking impedes the innate immune system by increasing mucus production and reducing mucociliary clearance, by damaging the epithelial barrier and stimulating the migration of various cells into the injured tissues. Furthermore, smoking stimulates the humoral and cellular components of the adaptive immune response to provide a more specific and diverse reaction that has an excellent memory for prior interaction between foreign material and the lung [7]. Figure 1 summarizes the inflammatory cell and mediator interactions involved in COPD pathogenesis. These are discussed below.

Macrophages are the most abundant cell type in bronchoalveolar lavage fluid (BALF) of COPD patients [2]. There is a five to tenfold increase in macrophage numbers in sputum, airways and lung parenchyma [11,12], and their numbers correlate with the severity of the disease [2]. Smoking and other noxious particles activate macrophages to produce tumor necrosis factor alpha (TNF-α), monocyte chemotactic peptide-1 (MCP-1), reactive oxygen species, and neutrophil chemotactic factors such as interleukin (IL)-8 and leukotriene (LT) B4, which are elevated in sputum from COPD patients [11]. Macrophages also secrete a wide array of tissue proteases, including matrix metalloproteinase (MMP)-1, MMP-2, MMP-9, MMP-12 and cathepsin K, L and S, which mediate the well-described lung parenchymal damage [4]. Alveolar macrophages (AMs) from COPD patients also show a distinct reduction in the ability to clean up apoptotic cells, with a resultant apoptotic cell build up [13]. In turn, this promotes more AM activation and further airway inflammation [14,15].

Sputum, BALF, bronchial biopsies and serum from COPD patients have elevated neutrophil numbers [11,12]. Chemoattractants such as IL-8, LTB4 and granulocyte macrophage-colony stimulating factor (GM-CSF) are involved in neutrophil mobilization into and prolonged survival in the lung interstitium; hence, their levels are increased in the airways of COPD patients [4,16–18]. Neutrophils are a rich source of inflammatory mediators including reactive oxygen species, lipid mediators and tissue proteases [4,19]. Importantly, neutrophil numbers in airways correlate not only to disease severity [11] but also to rate of decline in lung function [20].

Like neutrophils, $CD8^+$ T-cell numbers in the lungs of COPD patients are an excellent predictor of decline in lung function. In COPD lungs, they might contribute in the activation of macrophages to release neutrophilic chemotactic factors and proteases [7,19]. Furthermore, granzymes, which are effector protease molecules of $CD8^+$ T cells, have increased expression in type II pneumocytes in patients with severe COPD [21].

The inflammatory response in COPD has primarily been considered to be T-helper (TH)1 mediated, but recent developments in the understanding of cytokine biology have brought to light the presence of another discrete subpopulation of $CD4^+$ T cells, the TH17 phenotype [22]. TH17 cells might contribute to the inflammatory response through release of IL-17A and IL-17F, which are known to stimulate mesenchymal and bronchial epithelial cells to produce IL-6 [23] and to increase production of mucus via induction of MUC5AC and MUC5B genes in bronchial epithelial goblet cells and submucosal glands [24]. Moreover, IL-17s potently stimulate bronchial epithelial cells to generate CXCL8 (IL-8), a chemoattractant for neutrophils [25,26]. Of note, IL-17A is itself generated by IL-23, a cytokine of the innate immune system, suggesting the existence of a positive feedback loop [27,28]. This feedback loop involving IL-17 and IL-23 might explain the persistent and refractory inflammation in COPD despite smoking cessation. TH17 cells might be crucial in the development of COPD, and more work needs to be done to establish the function of these cells in the inflammatory pathways and potential of therapeutic options by inhibiting the IL-17–IL-23 feedback loops.

Cytokines and chemokines in COPD

Smoking and noxious airway irritants have been proposed to induce AMs, alveolar epithelial cells, T cells and dendritic cells to produce chemotactic factors for inflammatory cells. Cytokines and chemokines have been implicated in the pathogenesis of COPD, as discussed below [29] (Fig. 1).

Cytokines

TNF-α is an extremely potent pro-inflammatory mediator. There have been reports that TNF-α expression in patients with COPD is elevated, owing to either induction by cigarette smoke or genetic aberrations [30,31]. TNF-α has the capacity to induce inflammation not only by direct mechanisms but also by indirect ones. The activation of TNF-α receptor (TNFR) induces the production and release of several inflammatory mediators, including a broad spectrum of pro-inflammatory cytokines (GM-CSF, IL-1, TNF-α and IL-6), chemokines (CXCL8 and MCP-1), proteases (MMP-9 and MMP-12) [32–34] and adhesion molecules (intercellular adhesion molecule-1, or ICAM-1, and p-selectin) [35,36]. Furthermore, TNF-α has been implicated in various *in vitro* and *in vivo* studies in the induction of airway mucous cell metaplasia and hypersecretion [37], decreased interepithelial binding and cell death [38], emphysematous lesions and alveolar collagen deposition in murine alveolar walls [39], and induction of IFN-γ receptors on epithelial cells [40]. IFN-γ, in turn, inhibits the proliferation and decreases desmosome formation of epithelial cells [38]. In healthy smokers, sputum TNF-α levels were comparable to those of smoking-related COPD patients, but elevated neutrophil numbers and levels of soluble (s) TNFR-55 were noted in the diseased patients [41]. Furthermore, the sputum sTNFR correlated to the degree of airflow limitation. TNF-α might also be involved in the progression of COPD and disability because circulating TNF-α levels have been found to be inversely correlated with partial pressure of oxygen (pO_2) pO_2 and increased dyspnoea severity [42,43]. Moreover, TNF-α is involved systematically in 'inflammatory weight loss' [44]. Hence, antagonizing the effects of TNF-α, the multi-functional cytokine, might have a role in the treatment of COPD.

IL-6 is a potent pro-inflammatory mediator involved in the activation, growth, differentiation and survival of T cells [29,45]; in addition, in B cells, it is involved in antibody synthesis [46]. IL-6 levels are elevated in exhaled breath condensate, induced sputum, BALF and blood in COPD patients [47,48] during stable disease and increase further during exacerbations [49,50]. It is suggested that levels of IL-6 expression in BALF correlate to the severity of COPD [48]. On the basis of these reports, IL-6 inhibition might have a role in the treatment of COPD patients.

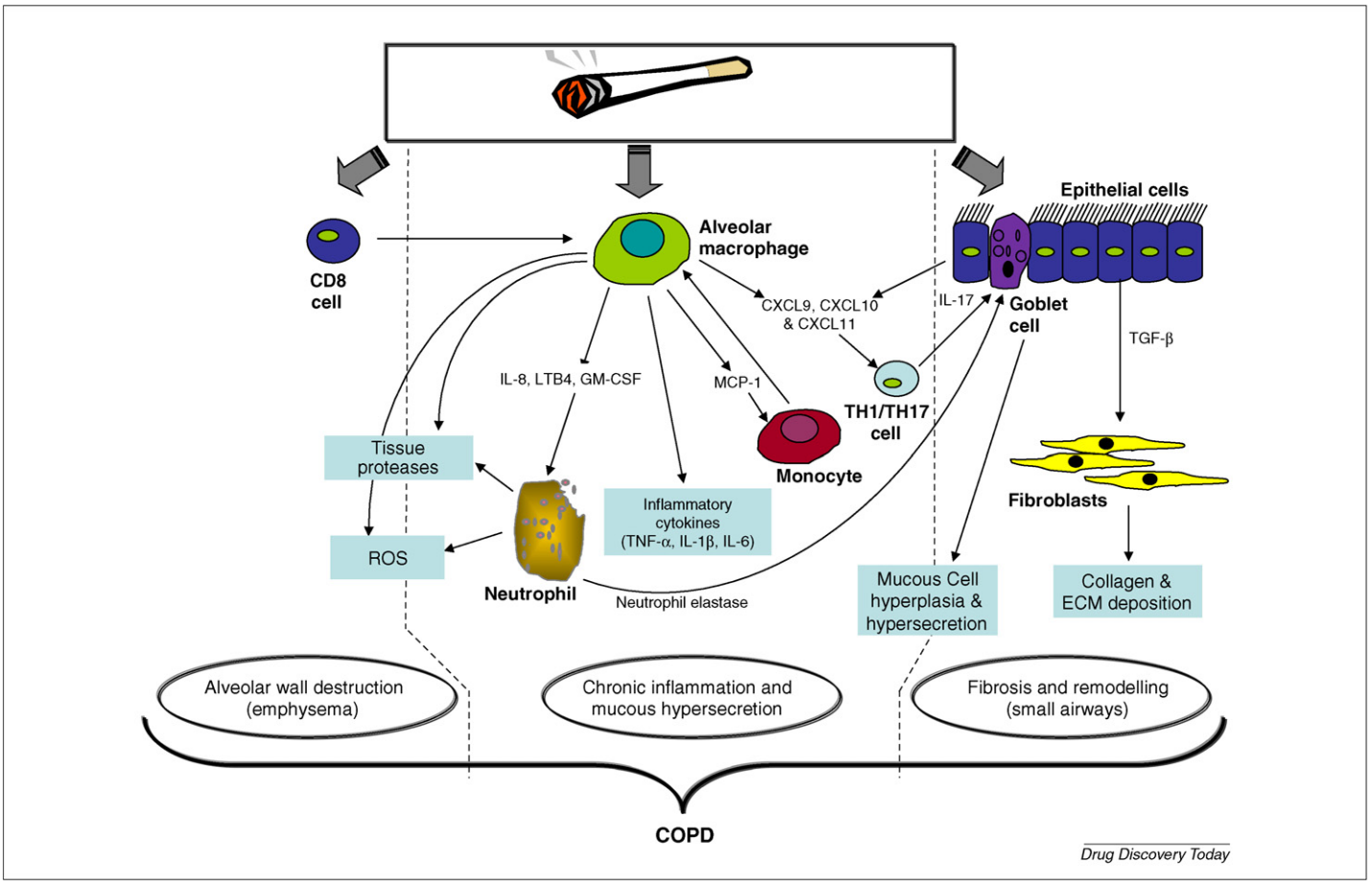

FIGURE 1

The inflammatory response to cigarette smoke and COPD pathogenesis. Cigarette smoke and other pollutants activate several inflammatory and structural cells of the lung to produce inflammatory and remodeling responses. AMs and neutrophils are the most abundant cell types in sputum, airways and lung parenchyma of COPD patients, and their numbers correlate with disease severity. When activated by cigarette smoke, AMs will produce a myriad of pro-inflammatory molecules (TNF-α, IL-1β and IL-6), chemokines (LTB4, CXCL8/IL-8, CCL2/MCP-1 and CXCL1/GRO-1α), ROS and tissue proteases (MMP-1, MMP-2, MMP-9, MMP-12 and cathepsin K, L and S), which mediate the well-described inflammatory response and lung damage in COPD. LTB4, CXCL8/IL-8 and GM-CSF secreted by AMs are also involved in the mobilization into and prolonged survival of neutrophils in the lung tissue. This cell type contributes to the inflammatory response and lung damage of COPD because they are a rich source of inflammatory mediators including ROS, lipid mediators and tissue proteases (neutrophil elastase and MMPs). The inflammatory response in COPD has primarily been considered to be $CD4^+$ TH1 mediated, but another discrete subpopulation of $CD4^+$ T cells has been characterized recently: the TH17 phenotype. TH17 cells might contribute to the inflammatory response and mucous hypersecretion through release of IL-17A and IL-17F, which are known to stimulate bronchial epithelial cells to produce IL-6 and CXCL8/IL-8 and to increase mucous production via induction of mucin genes in bronchial epithelial goblet cells and submucosal glands. In addition, $CD8^+$ T cells can be stimulated by cigarette smoke. They can contribute to alveolar wall destruction via the activation of AMs and their subsequent release of neutrophilic chemotactic factors and tissue proteases. Cigarette smoke might also directly activate alveolar epithelial cells to release further pro-inflammatory cytokines and TGF-β, which is known to modulate smooth muscle cell and fibroblast proliferation with subsequent progression to fibrosis and extracellular matrix deposition. *Abbreviations*: AMs, alveolar macrophages; GM-CSF, granulocyte macrophage-colony stimulating factor; GRO-1 α, growth-related oncogene 1; IL, interleukin; LTB4, leukotriene B4; MCP-1, monocyte chemotactic protein-1; MMP, matrix metalloproteinase; ROS, reactive oxygen species; TGF-β, transforming growth factor beta; TNF-α, tumour necrosis factor alpha.

Elevated levels of IL-1β have been measured in the sputum of stable COPD patients and correlate with neutrophil number, IL-8 and TNF-α [51]. Most of the evidence for an important role of IL-1β in COPD originated from work in rodents. Mice that overexpress IL-1β in lung epithelium during adulthood develop pulmonary inflammation, emphysema and airway remodeling [52]. Inhibition of IL-1β might have a role in COPD.

IL-10 is considered to be mainly an anti-inflammatory cytokine because it inhibits the synthesis of many inflammatory mediators and the activity of $CD4^+$ T cells [29]. In addition, IL-10 inhibits certain proteases (MMP-9) and increases the release of tissue inhibitors of MMPs, the endogenous inhibitors of MMPs [53]. Bronchial biopsies of COPD patients have increased expression of IL-10 [29]. Furthermore, $CD8^+$ T cells, which are known to synthesize IL-10, have been reported to be elevated in the BALF of patients with COPD compared to healthy smokers and non-smokers [54].

GM-CSF is a multi-functional pro-inflammatory cytokine produced by mesenchymal cells, endothelium, epithelial cells, monocytes and macrophages, as well as an autocrine mechanism [55]. Its functions include accelerated myelopoiesis; priming of leucocytes for activation and survival at inflammatory sites [56]; development of small airway fibrosis [57]; priming for sensitization to aeroallergens; direct inflammatory responses to the PP10 fraction (particles $<10\ \mu m$), which have been implicated in COPD exacerbations [58]; and driving the terminal differentiation of AMs by activating

the transcription factor PU-1 [59]. Concentrations of GM-CSF in BALF are elevated in stable disease and during exacerbations [60]. Smoking has been reported to upregulate GM-CSF mRNA [61]. Of note: although *in vitro* GM-CSF levels from COPD lung tissue are plentiful, its release is not antagonized by steroids [61]. Because glucocorticoids require intact HDAC, the reduced HDAC 2, 5 and 8 in COPD [62] might explain the glucocorticoid refractoriness observed in COPD. Murine model studies of subchronic exposure to cigarette smoke (four days) have shown an increase in inflammatory cells, cytokines, chemokines, proteases and toll-like receptors (TLRs) in BALF [55], and intrinsically administered GM-CSF antagonism to these mice significantly attenuated BALF macrophages, neutrophils and whole-lung cytokines, and chemokine and protease mRNA [61]. Antagonism of GM-CSF is also effective in experimental exacerbations of COPD triggered by bacterial surrogates and live viruses [61].

Chemokines and chemokine receptors

XC chemokines and receptors

CXCL8 (IL-8) and CXCL1 (growth-related oncogene-1) are expressed by mesenchymal, structural and inflammatory cells, and their expression can be stimulated by TNF-α, cigarette smoke and endotoxins. They are key mediators of neutrophil and monocyte chemotaxis and degranulation via the receptors CXCR1 and CXCR2 [29], and wound repair, angiogenesis, epithelial proliferation, endothelial migration and neovascularization via CXCR2 [63,64]. Levels of CXCL8 and CXCL1 are elevated in induced sputum and BALF in COPD patients compared with healthy smokers and non-smokers [11,16,48]. Although the use of antagonists to CXCR2 might impair repair and, hence, not be of much benefit, antagonism of CXCR1 is clearly valuable because by hampering neutrophil and monocyte chemotaxis, it will reduce the pro-inflammatory and destructive load from their mediators and proteases. In addition, direct CXCL8 blockage is currently exploited as a potential therapeutic target in COPD.

CC chemokines and receptors

Macrophages and monocytes express multiple chemokine receptors (CCR1, CCR2 and CCR5), which, when bound by ligands (MIP-α/CCL3, MIP-β/CCL4, MCP-1/CCL2, MCP-2/CCL8, MCP-3/CCL7 and MCP-4/CCL13), stimulate these cells to migrate *in vitro* [65–68]. *In vivo*, however, MCP-1 and CCR2 are important chemoattractants for the two cell types, and the former can also activate and attract mast cells and T cells [69,70]. CCR2 is the only known receptor for MCP-1 [71]. MCP-1 is synthesized by various cells, including AMs and mesenchymal and structural cells of the airways [72,73], and its expression is induced by other cytokines (TNF-α and IFN-γ) [34,74]. IFN-γ inhibits CCR2 expression [75], however, suggesting a probable natural inhibitory mechanism for the accumulation of macrophages. Multiple studies *in vivo* support distinct roles of MCP-1 and CCR2 in macrophage migration and the specificity of the MCP-1–CCR2 system rather than MIP-1α [65–68,76–78]. MCP-1 is also involved in the stimulation of endothelial wound healing by inducing endothelial migration [79], angiogenesis [80], vascular smooth muscle hyperplasia [81], collagen and TGF-β expression by fibroblasts [82], and the expression of adhesion molecules CD11c and CD11b, as well as IL-1 and IL-6, by blood monocytes [83]. Data confirm a pivotal role for MCP-1 and CCR2 in airway remodeling and inflammation directly or via macrophages/monocytes. Taking this into consideration, various antagonists of both MCP-1 and CCR2 have been developed and tested in murine models [68,84–86] with clinical and histological improvements in airway inflammation and hyperresponsiveness and bacterial clearance [66,77].

Clinical trials of novel anti-inflammatory agents in COPD

Current standard pharmacologic managements for COPD, which include short-acting and long-acting bronchodilators and inhaled corticosteroids (ICS), are simply directed at reducing symptoms and improving lung function but lack effective anti-inflammatory activity and fail to have an effect on disease progression. Long-acting β2-agonists (LABA) and muscarinic antagonists (LAMA) are both effective bronchodilators in patients with COPD. When administered together, the increase in lung function is significantly greater than for either alone [87]. This raises the concept of a LABA-LAMA combination as a 'super bronchodilator' for COPD. These drugs are currently in development. Another approach is a dual agent that has both M-3-antagonist and β2-agonist activity in a single molecule (MABA). ICS are largely ineffective in controlling the disease or improving physiological markers, but when combined with LABAs, they seem to acquire enhanced anti-inflammatory effects not seen with ICS alone [88–90]. Steroid insensitivity in COPD seems to be largely dependent on the reduced activity of histone deacetylase 2 (HDAC2), with low levels being reported in airway tissue from patients with COPD compared with healthy non-smokers [62]. It makes sense, therefore, to restore steroid sensitivity in COPD, and increasing HDAC2 expression and/or activation might be a potential approach. Low doses of oral theophylline have been shown to increase HDAC2 expression in AMs from COPD patients [91,92]. This novel molecular mechanism of action of theophylline, independent of PDE inhibition, has the potential to restore corticosteroid responsiveness *in vivo* and might explain some of the positive effects of the drug in COPD.

The need for effective reduction in pulmonary inflammation for patients with COPD has intensified the search for successful anti-inflammatory molecules, and this goal remains a key objective for academia and for the pharmaceutical industry. Some of these molecules have been assessed recently, and others are in the early stages of being evaluated. In the next section, we review the status of biologic and pharmacologic therapies that are currently in clinical development for COPD. Table 1 lists potential drugs that are currently in clinical development for COPD.

Anti-TNF-α drugs

As mentioned earlier, TNF-α has been implicated in the pathogenesis of COPD both locally and systemically. There have been three studies to assess the efficacy of TNF-α antagonism using the chimeric monoclonal antibody infliximab in COPD [93–95]. Infliximab manifests its activity by inhibiting the binding of soluble TNF-α to TNFRs, neutralizes soluble and membrane-bound TNF-α and has the potential to dissociate sTNFR complexes.

In the first study, van der Vaart *et al.* [93] performed an exploratory double-blind, placebo-controlled, randomized trial in 22 (14 infliximab; 8 placebo) current smokers with mild-to-moderate

TABLE 1

Selected biologic therapies in clinical development for COPD

Drug	Target	Biological/clinical effects	Adverse side-effects	Refs
Infliximab	TNF-α antagonist	No subjective or objective improvements shown in three large trials. Possibly useful in some selected COPD patients.	Increased incidence of respiratory tract malignancies and severe lung infections	[93–95]
ABX-IL-8	IL-8 (CXCL8) antagonist	Reduction of dyspnoea in a large RCT with three iv infusions. Efficacy is probably short-lived.	No major side-effects reported	[97]
ACZ-885	IL-1β antagonist	Inhibition of cigarette-smoke-induced pulmonary inflammation and emphysema in mice. A phase II RCT is ongoing to assess safety and efficacy.	No data available	[100]
SCH-527123 SB-656933 AZD-4818	Chemokine receptor (CXCR1/R2, CXCR2 and CCR1) antagonists	In preclinical models of COPD, inhibition of tissue infiltration of neutrophils/monocytes/T cells and attenuation of pulmonary inflammation/remodeling. Phase II RCTs with SCH-527123 are ongoing to assess safety and efficacy in smokers and in neutrophilic asthmatics.	No data available	[103,104,125, 101,102]
AZD-9668 ONO-6818	Neutrophil elastase inhibitors	ONO-6818 is currently evaluated at the preclinical stage. Phase II studies with the oral neutrophil elastase inhibitor (AZD 9668) are ongoing to evaluate safety and efficacy.	No data available	[105]
AZD-1236 AZD-3342 AZD-6067 Maristamat	MMP inhibitors	Most of these compounds prevented emphysema and small airways thickening in preclinical models of COPD. Clinical development has been stopped for many of these compounds.	Major side-effects (relevant toxicity)	[106–108]
SB-681323 GW 856553 PH 797804	p38 MAPK inhibitors	These compounds have shown attenuation of pulmonary inflammation/remodeling in several animal models of COPD. SB-856553 is in phase I trial, whereas a phase II RCT is ongoing to assess the anti-inflammatory activity, efficacy and safety of the oral p38 MAPK inhibitor 681323 in COPD.	Important side-effects and toxicity. Might be useful as an inhalatory delivery to reduce systemic exposure	[112–116,126]
R667	γ-Selective retinoid receptor antagonist	Two large RCTs (REPAIR and TEγSRA) are ongoing to assess safety and efficacy in COPD patients with emphysema.	Preliminary data confirmed safety of R667 for long-term use at doses up to 5 mg/day	[121–123]

CCR1, chemokine receptor; iv, intravenous; MAPK, mitogen-activated protein kinase; MCP-1, monocyte chemotactic protein-1; MMP, metalloproteinase; NF-κB, nuclear factor kappa B; RCT, randomized control trial; TGF-β, transforming growth factor beta.

COPD. Subjects received infusions of 5 mg/kg of infliximab or placebo at week 0, 2 and 6, where the primary outcome measure was % sputum neutrophil reduction besides various secondary endpoints of lung function, quality of life, bronchial hyperresponsiveness (BHR) and exhaled nitric oxide. It was reported that infliximab had no overall positive effect on the % sputum neutrophils, lung function, resting energy expenditure or quality of life. No serious adverse events or increase in respiratory infections were reported during nine weeks of follow-up.

In another small study, TNF-α antagonism using infliximab was assessed in COPD subjects with cachexia, which is characterized by inflammation reflected by elevated TNF-α levels [94]. Systemic inflammation and localized inflammation (using exhaled breath condensate) were evaluated. In this double-blind, placebo-controlled, randomized study, 41 (16 infliximab; 25 placebo) cachectic moderate-to-severe COPD patients were randomized to receive infliximab at 5 mg/kg or placebo at week 0, 2 and 6. Subjects were reviewed at week 8, 12 and 26. Although it was noted in the exhaled breath condensate (EBC) that there were elevated levels of MIP, IL-12, RANTES and sICAM-1 in cachectic COPD patients compared with placebo, treatment with infliximab had no effect on these markers. Furthermore, there were no changes in systemic acute-phase proteins (C-reactive protein (CRP), fibrinogen and lipopolysaccharide-binding protein), cytokine (IL-6) or sTNFR55. Hence, TNF-α blockade demonstrated no observable reduction in local or systemic markers of inflammation.

Finally, the clinical trial that was the longest and largest study of TNF-α blockade in COPD was conducted to determine the clinical benefit and safety in Global initiative for chronic Obstructive Disease (GOLD)-staged moderate-to-severe subjects [95]. This was a multicentre double-blind, placebo-controlled, randomized trial in which subjects were given infliximab at 3 mg/kg ($n = 78$) and 5 mg/kg ($n = 79$) or placebo ($n = 77$) at week 0, 2, 6, 12, 18 and 24. The primary endpoint was a change in the quality of life from baseline, as measured using the Chronic Respiratory Questionnaire, along with secondary endpoints of lung function, 6 min walk distance (6MWD) test, transition dyspnoea index, COPD exacerbations and SF-36 physical scores. Subjects were followed until week 44. Unfortunately, as in the other clinical trials of TNF-α antagonism in COPD, there were no significant improvements in primary or secondary endpoint assessed. Unlike in the study of Dentener *et al*. [94], however, a post hoc analysis reported that younger or cachectic COPD subjects at baseline had a significant increase in 6MWD as a result of treatment with infliximab compared with placebo. However, it was reported that 9 of 157 in the infliximab-treated compared with 1 of 77 placebo-treated subjects during the course of the trial were diagnosed with a malignancy. In addition, there was an increased incidence of pneumonias in the infliximab group compared with placebo, but no infection-related mortality was noted. There was a higher rate of study dropouts in the infliximab group (20–27%) compared with placebo (9%).

Overall, although there is considerable evidence suggesting that TNF-α has a role in COPD, administration of TNF-α blockade using infliximab in clinical trials up to six months has not shown any subjective or objective improvements. This raises the question of whether TNF-α is a pivotal cytokine in the pathogenesis of COPD. TNF-α might be useful in small subgroups of COPD patients and, hence, appropriate selection criteria of these subjects and outcome measures to assess the efficacy should be carefully chosen in future trials. Clearly, the use of infliximab routinely in moderate-to-severe COPD is not indicated. Besides, TNF-α antagonists in COPD and other inflammatory diseases, such as rheumatoid arthritis (RA) [96], have shown increased incidences of malignancies and severe infections, so the use of these agents needs careful evaluation not only for efficacy but also for safety issues.

IL-8 antagonism

IL-8 is responsible for developing chemotactic activity for neutrophils and $CD8^+$ cells and activates neutrophils by increasing degranulation and neutrophil elastase release. Mahler *et al*. examined the efficacy and safety of ABX-IL-8, a human monoclonal IgG2 antibody against IL-8 (CXCL8) in moderate-to-severe COPD subjects, all of whom had a component of chronic bronchitis [97]. This was a double-blind, placebo-controlled multicentre trial in which 109 (59 ABX-IL-8; 60 placebo) were enrolled to receive three intravenous infusions of either ABX-IL-8 (800 mg loading dose and 400 mg subsequent doses) or placebo over a three-month period. The primary outcome measure was a reduction in dyspnoea as measured by the transition dyspnoea index and secondary measures of health status including lung function, 6MWD and rescue inhaler use. It was observed that ABX-IL-8 reduced the severity of the dyspnoea, using the transition dyspnoea index, two weeks after the initial infusion and this persisted for a two-month period compared to placebo. However, there were no significant differences in lung function, health status or rescue inhaler use between the two groups. This pilot study showed that treatment with ABX-IL-8 is safe and well tolerated; the numbers of adverse events were similar in the two groups. Although the trial showed an improvement in the dyspnoea, the effect is probably short-lived because it was observed only at week 2 in their follow-up period. Further trials of IL-8 antagonism would probably be better via an inhalational agent with optimal doses and appropriate endpoints.

Other drug candidates in development

Pulmonary inflammation induced by multiple exposures to tobacco smoke in mice has been reported to be inhibited by treatment with an anti-IL-1β monoclonal antibody [98]. Furthermore, IL-1-receptor-knockout mice have significantly reduced emphysema after chronic exposure to tobacco smoke [99]. A monoclonal antibody against IL-1β (ACZ885) is currently under clinical development by Novartis. A phase II, randomized, double-blind and placebo-controlled study is ongoing to assess safety and efficacy of multiple doses of ACZ885 on pulmonary function in patients with COPD [100].

In addition, MCP-1 levels are elevated in the sputum from COPD patients and correlate with neutrophil number and lung function (FEV_1) [16]. Furthermore, MCP-1 and its receptor, CCR2, seem to be increased on macrophages and epithelial cells obtained from COPD patients [72]. In view of this, Novartis is seeking neutralization of MCP-1 in COPD by means of an anti-MCP-1 monoclonal antibody (ABN912). A similar compound, AZD4818 (a CCR1 antagonist that can be administered by inhalation), is being developed by AstraZeneca [101,102].

A novel potent antagonist of the chemokine receptor CXCR2, named SCH 527123, that inhibits neutrophil recruitment in several animal models of pulmonary inflammation is being developed

as an oral treatment to modify the progression of chronic inflammatory conditions such as COPD. The reversible decline in circulating neutrophils is probably because of the effects of SCH 527123 on neutrophil trafficking and/or neutrophil margination. A preliminary randomized, placebo-controlled, parallel-group study of 12 healthy smokers by Schering-Plough showed multiple-dose safety and tolerability of the compound [103]. Moreover, another phase II randomized study on safety of SCH 527123 in subjects with neutrophilic asthma has just been completed [104].

There is good evidence that an imbalance between naturally occurring proteases and anti-proteases that hold them in check is responsible for the tissue damage seen in late-stage COPD. Tissue proteases such as neutrophil elastase, MMPs and cathepsins are some of the known proteases released from activated neutrophils, macrophages and epithelial cells in COPD. Several protease inhibitors are in clinical development. Phase II studies by AstraZeneca are evaluating an oral neutrophil elastase inhibitor (AZD 9668) [105] and an oral MMP inhibitor (AZD 1236) [106–108]. AstraZeneca also has one candidate compound (AZD 3342) in phase I clinical trials and another in preclinical development (AZD 6067). In addition, Ono Pharmaceuticals has a neutrophil elastase inhibitor (ONO-6818) at the preclinical stage. In animal models, this was able to prevent changes typical of emphysema, such as airspace enlargement, loss of elastic recoil and lung hyperinflation. Moreover, MMP408, a potent and selective MMP-12 inhibitor, has been derived from a potent MMP-2 and -13 inhibitor via lead optimization and has good physical properties and bioavailability. The compound blocks rhMMP-12-induced lung inflammation in a mouse model and was advanced for further development for the treatment of COPD [109].

Instead of blocking the action of individual cytokines, other compounds could be designed to inhibit their synthesis, thereby blocking several inflammatory pathways simultaneously. Kinase-based signaling cascades have a crucial role in the expression and activation of inflammatory mediators in the airway and in T-cell function and airway remodeling by environmental stimuli such as tobacco smoke and by endogenous signals such as cytokines and inflammation-derived oxidants. Important kinases such as inhibitor of NF-κB kinase (IKK) 2, mitogen-activated protein kinases (MAPKs) and phosphoinositol-3 kinase (PI3K) regulate inflammation generally through activation of pro-inflammatory transcription factors such as activating protein-1 and nuclear factor kappa B (NF-κB), which are known to be activated in airway disease. Selective kinase inhibitors have been developed to reduce inflammation. One of the key regulators of inflammation is the enzyme p38 MAPK. This enzyme controls synthesis of IL-8, TNF-α and enzymes that are involved in tissue destruction in emphysema. The potential role of MAPK inhibitors in COPD therapy has been demonstrated in animal models [110]. A selective p38-α-kinase inhibitor, GW-856553, suppressed lipopolysaccaride (LPS)-induced p38 kinase activity in blood monocytes and inhibited IL-8 production in peripheral blood mononuclear cells from patients with COPD [111]. This compound also synergistically increased the suppressive effects of dexamethasone, the same inhibition of IL-8 being achieved with a significant reduction in steroid concentration [62]. GlaxoSmithKline has the p38 MAPK inhibitor GW-856553 in phase II trials [112–114] and has recently completed phase II studies with the oral p38 MAPK inhibitor SB-681323 to assess its anti-inflammatory activity, efficacy and safety in subjects with COPD [114–116].

There is evidence of significantly increased PI3K activity in peripheral blood monocytes from patients with COPD compared with smokers and normal control subjects. This is associated with decreased sensitivity to corticosteroids. The addition of a PI3K inhibitor restores steroid sensitivity toward normal, and other studies suggest that the delta isoform of PI3K is a potential target in restoring corticosteroid sensitivity in COPD [117]. There are several PI3K-delta inhibitors under development, including GSK373561A.

NF-κB regulates the expression of CXCL8 and other chemokines, TNF-α and other inflammatory cytokines, and MMP-9. NF-κB is activated in the macrophages and epithelial cells of COPD patients, especially during exacerbations. Small-molecule inhibitors of the inhibitor of IKK2 were tested, and an IKK2 inhibitor has been effective in some animal models of COPD [118]. Although several IKK2 inhibitors are now in development, at present none have been tested in COPD patients. IKK2 inhibitors are able to block the activation of NF-κB-activated genes and might the inhibit CXCR3 chemokines, indicating complex interactions between signal transduction pathways [119,120]. IKK2 inhibitors might be effective in suppressing the corticosteroid resistance inflammation of COPD. One concern about the long-term inhibition of NF-κB is that effective inhibitors might result in immune suppression and impair host defenses.

Among new compounds, R667 is an oral γ-selective retinoid receptor antagonist that in animal models of emphysema has been demonstrated to reduce inflammation and to promote structural repair and functional improvement. Human studies with R667 confirmed its safety for long-term use at doses up to 5 mg/day [121]. R667 is believed to be the only potential disease-modifying therapy in development for the treatment of emphysema. Two clinical studies are now evaluating its role in human emphysema treatment: the Retinoid Treatment of Emphysema in Patients on the Alpha 1-Antitrypsin International Registry (REPAIR) study has completed enrollment in Europe for 260 ex-smokers with emphysema to assess the safety and efficacy of R667 5 mg/day in a double-blind, placebo-controlled randomized study of one year [122]. The Treatment of Emphysema with γ-Selective Retinoid Agonist (TEγSRA) study is a two-year study that begun enrolling in the USA and EU/Africa 480 ex-smokers with moderate-to-severe emphysema to assess 5 mg orally daily or placebo, in addition to a long-acting bronchodilator and ICS optimal therapy regimen [123]. Recent data show that evolving safety profile of R667 5 mg/day is adequate to study continuation [124].

Concluding remarks

Even though understanding of the disease has improved considerably over the past 10–20 years, this is unlikely to be translated into a notable clinical advantage for the COPD sufferer in the near future. Indeed, because the human lung seems to lack much capacity for self-repair in adulthood, hopes of reversing the tissue damage seen in COPD seem remote with the drug arsenal presently at our disposal. Current standard pharmacological treatments for COPD, such as bronchodilators and corticosteroids, are largely ineffective in controlling the disease and reversing its progression. Early detection of the disease, prevention of acute exacerbations

and finding more effective ways of helping people to stop smoking will remain the most immediate goals of disease management for some time, therefore, with a focus on preserving as much lung function as possible and slowing the rate of loss of function as far as possible.

COPD medications of greater efficacy – particularly newer agents that treat the underlying inflammation and remodeling – are very much needed however. Several molecules with anti-inflammatory and antiprotease activity have already have been assessed, and others are in the early stages of being assessed. Medicines blocking the action of key cytokines such as TNF-α or IL-8 might reduce the inflammatory response in COPD, but blocking individual cytokines might not be enough to efficiently suppress inflammation, and there are also concerns about the long-term safety (i.e. the possibility of increased infections and malignancies) and the elevated costs of recombinant human monoclonals. To circumvent the problem of blocking the action of individual cytokines, inhibition of kinase-based signaling cascades has been exploited and selective kinase inhibitors have been developed to reduce the expression and activation of inflammatory mediators in COPD. Unfortunately, systemic administration of P38 MAPK inhibitors might cause side-effects and toxicity, and inhalatory formulations should be engineered to reduce systemic exposure. Other kinase inhibitors, such as inhibitors of IKK-2 and PI3K, might exhibit a much improved toxicity profile. Some existing medicines with anti-inflammatory activity, such as peroxisome proliferator-activated receptor γ agonists (currently used to treat diabetes) and statins (used to lower blood cholesterol) might be found to have some value in COPD, and studies in this direction are also required.

Future trials should not only look into the best method of administration, optimal dosage and duration of treatment but also use appropriate outcome measures, subject selection and the inclusion of biomarkers to determine the efficacy, as well as the alternative mechanism of action, of biological and pharmacological agents in COPD. When designing these trials, it is also important to consider that because stopping smoking is the only intervention capable of radically altering the speed of disease progress, it is imperative that new medications should be tested in the context of study populations no longer exposed to tobacco smoke to maximize chances of success.

Conflict of interest statement

R.P. has acted as a speaker for CV Therapeutics, Novartis, Merck, AstraZeneca, GSK and Roche. In addition, he is a consultant for CV Therapeutics, Duska Therapeutics and NeuroSearch.

References

1 Pauwels, R.A. *et al.* (2001) Global strategy for the diagnosis, management, and prevention of chronic obstructive pulmonary disease. NHLBI/WHO Global Initiative for Chronic Obstructive Lung Disease (GOLD) Workshop summary. *Am. J. Respir. Crit. Care Med.* 163, 1256–1276

2 MacNee, W. (2005) Pathogenesis of chronic obstructive pulmonary disease. *Proc. Am. Thorac. Soc.* 2, 258–266

3 Salvi, S.S. and Barnes, P.J. (2009) Chronic obstructive pulmonary disease in non-smokers. *Lancet* 374, 733–743

4 Barnes, P.J. *et al.* (2003) Chronic obstructive pulmonary disease: molecular and cellular mechanisms. *Eur. Respir. J.* 22, 672–688

5 Pauwels, R.A. *et al.* (1999) Long-term treatment with inhaled budesonide in persons with mild chronic obstructive pulmonary disease who continue smoking. European Respiratory Society Study on Chronic Obstructive Pulmonary Disease. *N. Engl. J. Med.* 340, 1948–1953

6 Culpitt, S.V. *et al.* (1999) Effect of high dose inhaled steroid on cells, cytokines, and proteases in induced sputum in chronic obstructive pulmonary disease. *Am. J. Respir. Crit. Care Med.* 160, 1635–1639

7 Hogg, J.C. *et al.* (2004) The nature of small-airway obstruction in chronic obstructive pulmonary disease. *N. Engl. J. Med.* 350, 2645–2653

8 Kumar, R. *et al.* (2005) *Acute and Chronic Inflammation.* Elsevier Saunders

9 Grashoff, W.F. *et al.* (1997) Chronic obstructive pulmonary disease: role of bronchiolar mast cells and macrophages. *Am. J. Pathol.* 151, 1785–1790

10 Knowles, M.R. and Boucher, R.C. (2002) Mucus clearance as a primary innate defense mechanism for mammalian airways. *J. Clin. Invest.* 109, 571–577

11 Keatings, V.M. *et al.* (1996) Differences in interleukin-8 and tumor necrosis factor-alpha in induced sputum from patients with chronic obstructive pulmonary disease or asthma. *Am. J. Respir. Crit. Care Med.* 153, 530–534

12 Peleman, R.A. *et al.* (1999) The cellular composition of induced sputum in chronic obstructive pulmonary disease. *Eur. Respir. J.* 13, 839–843

13 Vandivier, R.W. *et al.* (2006) Burying the dead: the impact of failed apoptotic cell removal (efferocytosis) on chronic inflammatory lung disease. *Chest* 129, 1673–1682

14 Johann, A.M. *et al.* (2006) Recognition of apoptotic cells by macrophages activates the peroxisome proliferator-activated receptor-gamma and attenuates the oxidative burst. *Cell Death Differ.* 13 (9), 1533–1540

15 Patel, V.A. *et al.* (2006) Apoptotic cells, at all stages of the death process, trigger characteristic signaling events that are divergent from and dominant over those triggered by necrotic cells: implications for the delayed clearance model of autoimmunity. *J. Biol. Chem.* 281, 4663–4670

16 Traves, S.L. *et al.* (2002) Increased levels of the chemokines GROalpha and MCP-1 in sputum samples from patients with COPD. *Thorax* 57, 590–595

17 Gomez-Cambronero, J. *et al.* (2003) Granulocyte-macrophage colony-stimulating factor is a chemoattractant cytokine for human neutrophils: involvement of the ribosomal p70 S6 kinase signaling pathway. *J. Immunol.* 171, 6846–6855

18 Tanino, M. *et al.* (2002) Increased levels of interleukin-8 in BAL fluid from smokers susceptible to pulmonary emphysema. *Thorax* 57, 405–411

19 Stockley, R.A. (1999) Neutrophils and protease/antiprotease imbalance. *Am. J. Respir. Crit. Care Med.* 160, S49–S52

20 Stanescu, D. *et al.* (1996) Airways obstruction, chronic expectoration, and rapid decline of FEV1 in smokers are associated with increased levels of sputum neutrophils. *Thorax* 51, 267–271

21 Vernooy, J.H. *et al.* (2002) Local and systemic inflammation in patients with chronic obstructive pulmonary disease: soluble tumor necrosis factor receptors are increased in sputum. *Am. J. Respir. Crit. Care Med.* 166, 1218–1224

22 Kikly, K. *et al.* (2006) The IL-23/Th(17) axis: therapeutic targets for autoimmune inflammation. *Curr. Opin. Immunol.* 18, 670–675

23 Molet, S. *et al.* (2001) IL-17 is increased in asthmatic airways and induces human bronchial fibroblasts to produce cytokines. *J. Allergy Clin. Immunol.* 108, 430–438

24 Chen, Y. *et al.* (2003) Stimulation of airway mucin gene expression by interleukin (IL)-17 through IL-6 paracrine/autocrine loop. *J. Biol. Chem.* 278, 17036–17043

25 Jones, C.E. and Chan, K. (2002) Interleukin-17 stimulates the expression of interleukin-8, growth-related oncogene-alpha, and granulocyte-colony-stimulating factor by human airway epithelial cells. *Am. J. Respir. Cell Mol. Biol.* 26, 748–753

26 Vanaudenaerde, B.M. *et al.* (2003) Interleukin-17 stimulates release of interleukin-8 by human airway smooth muscle cells *in vitro*: a potential role for interleukin-17 and airway smooth muscle cells in bronchiolitis obliterans syndrome. *J. Heart Lung Transplant* 22, 1280–1283

27 Langrish, C.L. *et al.* (2004) IL-12 and IL-23: master regulators of innate and adaptive immunity. *Immunol. Rev.* 202, 96–105

28 Aggarwal, S. *et al.* (2003) Interleukin-23 promotes a distinct CD4 T cell activation state characterized by the production of interleukin-17. *J. Biol. Chem.* 278, 1910–1914

29 Kim, V. *et al.* (2008) New concepts in the pathobiology of chronic obstructive pulmonary disease. *Proc. Am. Thorac. Soc.* 5, 478–485

30 Mio, T. *et al.* (1997) Cigarette smoke induces interleukin-8 release from human bronchial epithelial cells. *Am. J. Respir. Crit. Care Med.* 155, 1770–1776

31 Sakao, S. *et al.* (2001) Association of tumor necrosis factor alpha gene promoter polymorphism with the presence of chronic obstructive pulmonary disease. *Am. J. Respir. Crit. Care Med.* 163, 420–422

32 Cromwell, O. *et al.* (1992) Expression and generation of interleukin-8 IL-6 and granulocyte-macrophage colony-stimulating factor by bronchial epithelial cells and enhancement by IL-1β and tumour necrosis factor-alpha. *Immunology* 77, 330–337

33 von Asmuth, E.J. *et al.* (1994) IL-6 IL-8 and TNF production by cytokine and lipopolysaccharide-stimulated human renal cortical epithelial cells in vitro. *Eur. Cytokine Netw.* 5, 301–310

34 Standiford, T.J. *et al.* (1991) Alveolar macrophage-derived cytokines induce monocyte chemoattractant protein-1 expression from human pulmonary type II-like epithelial cells. *J. Biol. Chem.* 266, 9912–9918

35 Di Stefano, A. *et al.* (1994) Upregulation of adhesion molecules in the bronchial mucosa of subjects with chronic obstructive bronchitis. *Am. J. Respir. Crit. Care Med.* 149, 803–810

36 Mulligan, M.S. *et al.* (1992) Neutrophil-dependent acute lung injury. Requirement for P-selectin (GMP-140). *J. Clin. Invest.* 90, 1600–1607

37 Takeyama, K. *et al.* (2001) Activation of epidermal growth factor receptors is responsible for mucin synthesis induced by cigarette smoke. *Am. J. Physiol. Lung Cell. Mol. Physiol.* 280, L165–L172

38 Kampf, C. *et al.* (1999) Effects of TNF-α. IFN-γ and IL-beta on normal human bronchial epithelial cells. *Eur. Respir. J.* 14, 84–91

39 Miyazaki, Y. *et al.* (1995) Expression of a tumor necrosis factor-alpha transgene in murine lung causes lymphocytic and fibrosing alveolitis. A mouse model of progressive pulmonary fibrosis. *J. Clin. Invest.* 96, 250–259

40 Wu, A.J. *et al.* (1996) Interferon-γ induced cell death in a cultured human salivary gland cell line. *J. Cell. Physiol.* 167, 297–304

41 Vernooy, J.H. *et al.* (2007) Increased granzyme A expression in type II pneumocytes of patients with severe chronic obstructive pulmonary disease. *Am. J. Respir. Crit. Care Med.* 175, 464–472

42 Takabatake, N. *et al.* (2000) The relationship between chronic hypoxemia and activation of the tumor necrosis factor-alpha system in patients with chronic obstructive pulmonary disease. *Am. J. Respir. Crit. Care Med.* 161, 1179–1184

43 Garrod, R. *et al.* (2007) The relationship between inflammatory markers and disability in chronic obstructive pulmonary disease (COPD). *Prim. Care Respir. J.* 16, 236–240

44 de Godoy, I. *et al.* (1996) Elevated TNF-α production by peripheral blood monocytes of weight-losing COPD patients. *Am. J. Respir. Crit. Care Med.* 153, 633–637

45 Park, J.Y. and Pillinger, M.H. (2007) Interleukin-6 in the pathogenesis of rheumatoid arthritis. *Bull. NYU Hosp. Jt. Dis.* 65 (Suppl. 1), S4–S10

46 Muraguchi, A. *et al.* (1988) The essential role of B cell stimulatory factor 2 (BSF-2/IL-6) for the terminal differentiation of B cells. *J. Exp. Med.* 167, 332–344

47 Man, S.F. *et al.* (2006) C-reactive protein and mortality in mild to moderate chronic obstructive pulmonary disease. *Thorax* 61, 849–853

48 Soler, N. *et al.* (1999) Airway inflammation and bronchial microbial patterns in patients with stable chronic obstructive pulmonary disease. *Eur. Respir. J.* 14, 1015–1022

49 Wedzicha, J.A. *et al.* (2000) Acute exacerbations of chronic obstructive pulmonary disease are accompanied by elevations of plasma fibrinogen and serum IL-6 levels. *Thromb. Haemost.* 84, 210–215

50 Bucchioni, E. *et al.* (2003) High levels of interleukin-6 in the exhaled breath condensate of patients with COPD. *Respir. Med.* 97, 1299–1302

51 Newbold, P. *et al.* (2005) Evidence to support a role for IL-1β in the COPD disease process. *Proc. Am. Thorac. Soc.* 2, A395

52 Lappalainen, U. *et al.* (2005) Interleukin-1beta causes pulmonary inflammation, emphysema, and airway remodeling in the adult murine lung. *Am. J. Respir. Cell Mol. Biol.* 32, 311–318

53 Lacraz, S. *et al.* (1995) IL-10 inhibits metalloproteinase and stimulates TIMP-1 production in human mononuclear phagocytes. *J. Clin. Invest.* 96, 2304–2310

54 Barcelo, B. *et al.* (2006) Intracellular cytokine profile of T lymphocytes in patients with chronic obstructive pulmonary disease. *Clin. Exp. Immunol.* 145, 474–479

55 Vlahos, R. *et al.* (2006) Differential protease, innate immunity, and NF-κB induction profiles during lung inflammation induced by subchronic cigarette smoke exposure in mice. *Am. J. Physiol. Lung Cell. Mol. Physiol.* 290, L931–L945

56 Hamilton, J.A. and Anderson, G.P. (2004) GM-CSF biology. *Growth Factors* 22, 225–231

57 Xing, Z. *et al.* (1996) Transfer of granulocyte-macrophage colony-stimulating factor gene to rat lung induces eosinophilia, monocytosis, and fibrotic reactions. *J. Clin. Invest.* 97, 1102–1110

58 Stampfli, M.R. *et al.* (1998) GM-CSF transgene expression in the airway allows aerosolized ovalbumin to induce allergic sensitization in mice. *J. Clin. Invest.* 102, 1704–1714

59 Shibata, Y. *et al.* (2001) GM-CSF regulates alveolar macrophage differentiation and innate immunity in the lung through PU.1. *Immunity* 15, 557–567

60 Balbi, B. *et al.* (1997) Increased bronchoalveolar granulocytes and granulocyte/macrophage colony-stimulating factor during exacerbations of chronic bronchitis. *Eur. Respir. J.* 10, 846–850

61 Vlahos, R. *et al.* (2006) Therapeutic potential of treating chronic obstructive pulmonary disease (COPD) by neutralising granulocyte macrophage-colony stimulating factor (GM-CSF). *Pharmacol. Ther.* 112, 106–115

62 Ito, K. *et al.* (2005) Decreased histone deacetylase activity in chronic obstructive pulmonary disease. *N. Engl. J. Med.* 352, 1967–1976

63 Strieter, R.M. *et al.* (1992) Interleukin-8. A corneal factor that induces neovascularization. *Am. J. Pathol.* 141, 1279–1284

64 Arenberg, D.A. *et al.* (1996) Inhibition of interleukin-8 reduces tumorigenesis of human non-small cell lung cancer in SCID mice. *J. Clin. Invest.* 97, 2792–2802

65 Boring, L. *et al.* (1997) Impaired monocyte migration and reduced type 1 (Th1) cytokine responses in C-C chemokine receptor 2 knockout mice. *J. Clin. Invest.* 100, 2552–2561

66 Kurihara, T. *et al.* (1997) Defects in macrophage recruitment and host defense in mice lacking the CCR2 chemokine receptor. *J. Exp. Med.* 186, 1757–1762

67 Lu, B. *et al.* (1998) Abnormalities in monocyte recruitment and cytokine expression in monocyte chemoattractant protein 1-deficient mice. *J. Exp. Med.* 187, 601–608

68 Mack, M. *et al.* (2001) Expression and characterization of the chemokine receptors CCR2 and CCR5 in mice. *J. Immunol.* 166, 4697–4704

69 Conti, P. *et al.* (1995) Monocyte chemotactic protein-1 provokes mast cell aggregation and [^{3}H]5HT release. *Immunology* 86, 434–440

70 Taub, D.D. *et al.* (1995) Monocyte chemotactic protein-1 (MCP-1), -2, and -3 are chemotactic for human T lymphocytes. *J. Clin. Invest.* 95, 1370–1376

71 Schweickart, V.L. *et al.* (2001) CCR11 is a functional receptor for the monocyte chemoattractant protein family of chemokines. *J. Biol. Chem.* 276, 856

72 de Boer, W.I. *et al.* (2000) Monocyte chemoattractant protein 1, interleukin 8, and chronic airways inflammation in COPD. *J. Pathol.* 190, 619–626

73 Rolfe, M.W. *et al.* (1992) Expression and regulation of human pulmonary fibroblast-derived monocyte chemotactic peptide-1. *Am. J. Physiol.* 263, L536–L545

74 Warhurst, A.C. *et al.* (1998) Interferon γ induces differential upregulation of alpha and beta chemokine secretion in colonic epithelial cell lines. *Gut* 42, 208–213

75 Penton-Rol, G. *et al.* (1998) Selective inhibition of expression of the chemokine receptor CCR2 in human monocytes by IFN-γ. *J. Immunol.* 160, 3869–3873

76 Gunn, M.D. *et al.* (1997) Monocyte chemoattractant protein-1 is sufficient for the chemotaxis of monocytes and lymphocytes in transgenic mice but requires an additional stimulus for inflammatory activation. *J. Immunol.* 158, 376–383

77 Gonzalo, J.A. *et al.* (1998) The coordinated action of CC chemokines in the lung orchestrates allergic inflammation and airway hyperresponsiveness. *J. Exp. Med.* 188, 157–167

78 Hautamaki, R.D. *et al.* (1997) Requirement for macrophage elastase for cigarette smoke-induced emphysema in mice. *Science* 277, 2002–2004

79 Weber, K.S. *et al.* (1999) Expression of CCR2 by endothelial cells: implications for MCP-1 mediated wound injury repair and *in vivo* inflammatory activation of endothelium. *Arterioscler. Thromb. Vasc. Biol.* 19, 2085–2093

80 Salcedo, R. *et al.* (2000) Human endothelial cells express CCR2 and respond to MCP-1: direct role of MCP-1 in angiogenesis and tumor progression. *Blood* 96, 34–40

81 Furukawa, Y. *et al.* (1999) Anti-monocyte chemoattractant protein-1/monocyte chemotactic and activating factor antibody inhibits neointimal hyperplasia in injured rat carotid arteries. *Circ. Res.* 84, 306–314

82 Gharaee-Kermani, M. *et al.* (1996) Costimulation of fibroblast collagen and transforming growth factor beta1 gene expression by monocyte chemoattractant protein-1 via specific receptors. *J. Biol. Chem.* 271, 17779–17784

83 Jiang, Y. *et al.* (1992) Monocyte chemoattractant protein-1 regulates adhesion molecule expression and cytokine production in human monocytes. *J. Immunol.* 148, 2423–2428

84 Gong, J.H. and Clark-Lewis, I. (1995) Antagonists of monocyte chemoattractant protein 1 identified by modification of functionally critical NH2-terminal residues. *J. Exp. Med.* 181, 631–640

85 Mirzadegan, T. *et al.* (2000) Identification of the binding site for a novel class of CCR2b chemokine receptor antagonists: binding to a common chemokine receptor motif within the helical bundle. *J. Biol. Chem.* 275, 25562–25571

86 Rodriguez-Frade, J.M. *et al.* (1999) The chemokine monocyte chemoattractant protein-1 induces functional responses through dimerization of its receptor CCR2. *Proc. Natl. Acad. Sci. U. S. A.* 96, 3628–3633

87 van Noord, J.A. *et al.* (2005) Comparison of tiotropium once daily, formoterol twice daily and both combined once daily in patients with COPD. *Eur. Respir. J.* 26, 214–222

88 Barnes, N.C. *et al.* (2006) Antiinflammatory effects of salmeterol/fluticasone propionate in chronic obstructive lung disease. *Am. J. Respir. Crit. Care Med.* 173, 736–743

89 Bourbeau, J. *et al.* (2007) Effect of salmeterol/fluticasone propionate on airway inflammation in COPD: a randomised controlled trial. *Thorax* 62, 938–943

90 Chung, K.F. *et al.* (2009) Inhaled corticosteroids as combination therapy with beta-adrenergic agonists in airways disease: present and future. *Eur. J. Clin. Pharmacol.* 65, 853–871

91 Cosio, B.G. *et al.* (2004) Theophylline restores histone deacetylase activity and steroid responses in COPD macrophages. *J. Exp. Med.* 200, 689–695

92 Ito, K. *et al.* (2002) A molecular mechanism of action of theophylline: induction of histone deacetylase activity to decrease inflammatory gene expression. *Proc. Natl. Acad. Sci. U. S. A.* 99, 8921–8926

93 van der Vaart, H. *et al.* (2005) First study of infliximab treatment in patients with chronic obstructive pulmonary disease. *Am. J. Respir. Crit. Care Med.* 172, 465–469

94 Dentener, M.A. *et al.* (2008) Effect of infliximab on local and systemic inflammation in chronic obstructive pulmonary disease: a pilot study. *Respiration*

95 Rennard, S.I. *et al.* (2007) The safety and efficacy of infliximab in moderate to severe chronic obstructive pulmonary disease. *Am. J. Respir. Crit. Care Med.* 175, 926–934

96 Bongartz, T. *et al.* (2006) Anti-TNF antibody therapy in rheumatoid arthritis and the risk of serious infections and malignancies: systematic review and meta-analysis of rare harmful effects in randomized controlled trials. *J. Am. Med. Assoc.* 295, 2275–2285

97 Mahler, D.A. *et al.* (2004) Efficacy and safety of a monoclonal antibody recognizing interleukin-8 in COPD: a pilot study. *Chest* 126, 926–934

98 Castro, P. *et al.* (2004) Inhibition of interleukin-1beta reduces mouse lung inflammation induced by exposure to cigarette smoke. *Eur. J. Pharmacol.* 498, 279–286

99 Churg, A. *et al.* (2009) The role of interleukin-1β in murine cigarette smoke-induced emphysema and small airway remodeling. *Am. J. Respir. Cell Mol. Biol.* 40 (4), 482–490

100 Safety and efficacy of multiple doses of ACZ885 in chronic obstructive pulmonary disease (COPD) patients. (http://www.clinicaltrial.gov/ct2/show/NCT00581945?term=COPD+AND+ACZ&rank=1)

101 Tolerability/safety and efficacy on inhaled AZD4818 in patients with moderate to severe chronic obstructive pulmonary disease (COPD) (TOP). http://www.clinicaltrial.gov/ct2/show/NCT00629239?term=AZD-4818&rank=2

102 Norman, P. (2009) AZD-4818, a chemokine CCR1 antagonist: WO2008103126 and WO2009011653. *Expert Opin. Ther. Pat.* 19, 1629–1633

103 Study of the effects of SCH 527123 in subjects with moderate to severe chronic obstructive disease (COPD) (Protocol No. P05575). http://www.clinicaltrial.gov/ct2/show/NCT01006616?term=sch-527123&rank=6

104 Neutrophilic asthma study with SCH 527123 (Study P05365AM2). http://www.clinicaltrial.gov/ct2/show/NCT00632502?term=sch-527123&rank=5

105 Efficacy and safety of twice daily 60 mg AZD9668 in COPD for 12 weeks in patients on background budesonide/formoterol. http://www.clinicaltrial.gov/ct2/show/NCT01023516?term=AZD-9668+COPD&rank=3

106 Safety/tolerability study with AZD1236 in chronic obstructive pulmonary disease (COPD) patients (CERA). http://www.clinicaltrial.gov/ct2/show/NCT00758459?term=AZD1236+copd&rank=1

107 A phase IIa study assessing the effects of AZD1236 on biomarkers in chronic obstructive pulmonary disease (COPD) patients (BICO). http://www.clinicaltrial.gov/ct2/show/NCT00758706?term=AZD1236+copd&rank=2

108 Norman, P. (2009) Selective MMP-12 inhibitors: WO-2008057254. *Expert Opin. Ther. Pat.* 19, 1029–1034

109 Li, W. *et al.* (2009) A selective matrix metalloprotease 12 inhibitor for potential treatment of chronic obstructive pulmonary disease (COPD): discovery of (*S*)-2-(8-(methoxycarbonylamino)dibenzo[*b*,*d*]furan-3-sulfonamido)-3-methylbutanoic acid (MMP408). *J. Med. Chem.* 52, 1799–1802

110 Medicherla, S. *et al.* (2008) p38alpha-selective mitogen-activated protein kinase inhibitor SD-282 reduces inflammation in a subchronic model of tobacco smoke-induced airway inflammation. *J. Pharmacol. Exp. Ther.* 324, 921–929

111 Bhavsar, P.K. *et al.* (2009) Reversal of relative corticosteroid insensitivity in PBMCs from patients with COPD by p38 MAPK inhibition. *Am. J. Respir. Crit. Care Med.* 179, A6187

112 Safety and anti-inflammatory effect of SB681323 in patients with chronic obstructive pulmonary disease (COPD). http://www.clinicaltrial.gov/ct2/show/NCT00144859?term=681323+COPD&rank=2

113 Singh, D. *et al.* (2010) A randomized, placebo-controlled study of the effects of the p38 MAPK inhibitor SB-681323 on blood biomarkers of inflammation in COPD patients. *J. Clin. Pharmacol.* 50, 94–100

114 Bourbeau, J. and Johnson, M. (2009) New and controversial therapies for chronic obstructive pulmonary disease. *Proc. Am. Thorac. Soc.* 6, 553–554

115 A 12 week study to assess efficacy and safety of GW856553 in subjects with chronic obstructive pulmonary disease (COPD). http://www.clinicaltrial.gov/ct2/show/NCT00642148?term=856553+COPD&rank=1

116 A study to investigate the effects of GW856553 on patients with COPD (chronic obstructive pulmonary disease). http://www.clinicaltrial.gov/ct2/show/NCT00392587?term=856553+COPD&rank=2

117 Marwick, J.A. *et al.* (2009) Inhibition of PI3Kdelta restores glucocorticoid function in smoking-induced airway inflammation in mice. *Am. J. Respir. Crit. Care Med.* 179, 542–548

118 Birrell, M.A. *et al.* (2006) IkappaB kinase-2-independent and -dependent inflammation in airway disease models: relevance of IKK-2 inhibition to the clinic. *Mol. Pharmacol.* 69, 1791–1800

119 Tudhope, S.J. *et al.* (2007) The role of IkappaB kinase 2, but not activation of NF-κB, in the release of CXCR3 ligands from IFN-γ-stimulated human bronchial epithelial cells. *J. Immunol.* 179, 6237–6245

120 Bamborough, P. *et al.* (2009) Progress towards the development of anti-inflammatory inhibitors of IKKbeta. *Curr. Top. Med. Chem.* 9, 623–639

121 Roth, M.D. *et al.* (2006) Feasibility of retinoids for the treatment of emphysema study. *Chest* 130, 1334–1345

122 Repair Tier 2 – (retinoids in emphysema patients on the Alpha-1-Antitrypsin International Registry) (Protocol No NA17598). http://www.roche-trials.com/patient/trials/trial102.html

123 TESRA (treatment of emphysema with a gamma-selective retinoid agonist). http://www.clinicaltrial.gov/ct2/show/NCT00413205?term=emphysema+AND+retinoid&rank=2

124 Kvale, P. *et al.* (2007) Design of two studies in emphysema with an orally active, gamma selective retinoid agonist (R667). *Chest* 132, 481

125 A study to evaluate the safety and tolerability of SB-656933-AAA following repeated doses in healthy adult subjects. http://www.clinicaltrial.gov/ct2/show/NCT00504439?term=CXCR+copd&rank=1

126 A phase II study to evaluate the efficacy and safety of ph-797804 in adults with moderate to severe chronic obstructive pulmonary disease (COPD). http://www.clinicaltrial.gov/ct2/show/NCT00559910?term=797804+copd&rank=1