As chronic obstructive pulmonary disease cannot be treated effectively by corticosteroids, what are the perspectives of specific cytokine antagonists for therapy?

Perspectives for cytokine antagonist therapy in COPD

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Chronic obstructive pulmonary disease (COPD) is an inflammatory lung disease associated with progressive airflow limitation. The main risk factor is tobacco smoking. Anti-inflammatory therapies (e.g. corticosteroids) are based on those developed initially for asthma. In contrast to asthma, they are rather ineffective in improving chronic symptoms and reducing inflammation and lung function decline. Specific drugs need to be developed directed against this chronic inflammation, thereby preventing lung tissue damage. Cytokine antagonists for tumour necrosis factor α, interleukin 8 receptor, interleukin 1, and specific signal transduction inhibitors have proven to be effective for other inflammatory diseases. Their efficacy for COPD therapy has yet to be demonstrated.
factor (TNF) α, matrix metalloproteinase or antioxidant genes [5–10] as potential susceptibility factors dependent upon race and COPD phenotype [10].

Pathology and pathophysiology
COPD can be divided into three different disorders: chronic bronchitis with airflow limitation, emphysema and chronic bronchiolitis, each of which has its own clinical or pathological definition. COPD is characterised by a chronic airway inflammation and a progressive decline in lung function, measured as an increased annual decline in forced expiratory flow in one second (FEV₁) [1,2]. Systemic consequences, including systemic inflammation and declines in skeletal muscle mass and function are also seen [11–13]. In emphysema, alveolar seps are deteriorated resulting in reduced gas exchange, and loss of elastic recoil of lung tissue [14]. In patients with chronic bronchitis, bronchial gland enlargement and goblet cell hyperplasia lead to an overproduction of mucus, resulting in obstruction of mucoid airways [14]. Often, patients show a combination of these disorders. Research in the past two decades showed pathological features of COPD patients comprising lung tissue remodelling-like changes in mucosal tissue fibre types and/or fibrosis, pulmonary and systemic inflammation, and lung vascular remodelling [12,13–19].

The pulmonary inflammation consists of neutrophils, macrophages, CD8+ T cells (but less prominently in severe COPD) mast cells, and also eosinophils during exacerbations or in COPD patients showing reversible lung function [17,20,21]. Enhanced extrapulmonary inflammation has been noted including higher CD8+ T cell numbers (nasal mucosa, paratracheal lymph nodes) or neutrophils (nasal mucosa) in smokers with COPD compared with smokers without COPD or non-smokers [18,19].

Pathophysiological features in addition to decreased lung function include chronic cough and dyspnoea. In smokers, epithelial cell metaplasia often occurs, resulting in chronically decreased ciliary function. In chronic bronchitis, the hypersecreted mucus cannot be cleared sufficiently, resulting in chronic cough and dyspnoea. Whereas

| FIGURE 1 | Distribution and expression of chemokines and their receptors in human lungs. B lymphocytes and plasma cells express a plethora of chemokines and receptors in a tissue- and differentiation-dependent manner. However, the chemokine expression profile of human lung plasma cells has not yet been published. The proteins in bold are considered to be major molecules in the pathophysiology of COPD, and are discussed as potential therapeutic targets. Abbreviations: mφ, monocyte/macrophage; nφ, neutrophil; T, T cell; eos, eosinophil; MC, mast cell; DC, dendritic cell; plasma, plasma cell; epi, epithelial cell; SM, smooth muscle cell; endo, endothelial cell; fibro, fibroblast. Ref [21]. |
after stopping smoking the decline in lung function is reduced, lung function will not be restored and increased airway resistance persists [22–25]. This is probably due to pathological alterations like fibrosis and inflammation that reduce the diameter of airway lumen. In addition, after stopping smoking the inflammation persists, hence contributing to the irreversibility of the decreased lung function [25]. Exacerbations occur frequently in COPD patients and can eventually result in hospitalisation. These patients show increased airway inflammation with eosinophils and neutrophils and higher levels of inflammatory mediators like cytokines [26–28]. Exacerbations are thought to be caused by bacterial and viral infections [28]. As CD8+ T cells are involved in resolving viral infections, the lower numbers of these cells in patients with severe COPD may explain the frequent incidence of viral infections in these patients [20].

Other disturbed mechanisms are thought to include oxidative stress and protease-antiprotease balance [12,15,17,21,29]. This could be due to genetic predisposition, as seen in patients with α1-antitrypsin deficiency, or due to chronic exposition to cigarette smoke and subsequent release of free radicals and proteases like elastase and matrix metalloproteinases (MMPs) from activated inflammatory cells. In turn, this can cause a chronic state of tissue damage, repair and remodelling.

**Inflammation, cytokines and chemokines**

Inflammatory cells of the innate immune system seem to play an important role in the pathophysiology of COPD [14]. Pulmonary inflammation with macrophages and CD8+ T cells was demonstrated in airway walls and vascular smooth muscle, and neutrophils in bronchial glands and the airway lumen [17,21]. These cells migrate to chemotactic gradients of, and are activated by, small proteins: cytokines and chemokines. Classical cytokines include interleukins, interferons and TNFα. Based on structural homology, chemokines can be subdivided into four subclasses, of which CXC (or α) and CC (or β) chemokines account for most of them [21]. Cytokines and chemokines are synthesized by structural cells (such as fibroblasts, epithelial, endothelial, and muscle cells) and inflammatory cells. They act via specific membrane-bound receptors resulting in cell-specific reactions. Figure 1 gives an overview of the expression of the main chemokines and their receptors by the different cells in the lungs. With regard to COPD, protein and/or mRNA levels of different cytokines and chemokines have been found to be altered compared with subjects without COPD (Table 1). Among these, TNFα or TNFR levels, soluble IL-1 receptor antagonist (sIL-1Ra), CCL2 (or monocyte chemotactrant protein 1, MCP-1) and its receptor CCR2, CCL3 (or macrophage inflammatory protein 1α, MIP-1α) and CCL4 (or MIP-1β) and their receptor CCR5, CXCL8 (or IL-8), and CXCL10 (or interferon-inducible protein 10, IP-10) can be discerned as pro-inflammatory factors. In Figure 2, a brief outline of the proposed inflammatory mechanism in COPD is provided [21,30]. Based on this, anti-inflammatory molecules have therapeutic potential for COPD.

**Current therapies**

Current therapies for COPD are mainly based on drugs that were aimed at reducing pulmonary inflammation in asthma. These include oral or inhaled corticosteroids with or without bronchodilators including β2-agonists [1,31; GOLD guidelines: www.goldcopd.com]. Some COPD studies reported reduction of the number of exacerbations and improved quality of life [1,31,32], improved decline in FEV1 after short- or long-term treatment [33,34], or no effect on lung function [35] upon inhaled corticosteroid treatment. In contrast to asthma and COPD patients with bronchial hyperresponsiveness and eosinophilia, steroid treatment in COPD is rather ineffective in reducing airway

**TABLE 1**

Expression and polymorphisms of cytokines and chemokines in COPD

<table>
<thead>
<tr>
<th>Cytokine</th>
<th>Product</th>
<th>Compartment</th>
<th>Data</th>
<th>Refs</th>
</tr>
</thead>
<tbody>
<tr>
<td>TNFα</td>
<td>Protein</td>
<td>ser, sp, leg muscle</td>
<td>Higher [17,30]</td>
<td></td>
</tr>
<tr>
<td>mRNA</td>
<td>Protein</td>
<td>ser, sp</td>
<td>Higher [17,30]</td>
<td></td>
</tr>
<tr>
<td>Polymorph</td>
<td>Gene</td>
<td>Yes</td>
<td>Higher [17,30]</td>
<td></td>
</tr>
<tr>
<td>sTNFR p55</td>
<td>Protein</td>
<td>sp</td>
<td>Higher [17,30]</td>
<td></td>
</tr>
<tr>
<td>sTNFR p75</td>
<td>Protein</td>
<td>ser</td>
<td>Higher [17,30]</td>
<td></td>
</tr>
<tr>
<td>sIL-1Ra</td>
<td>Protein</td>
<td>ser</td>
<td>Higher [17,30,50]</td>
<td></td>
</tr>
<tr>
<td>IL-6</td>
<td>Protein</td>
<td>ebc</td>
<td>Higher [52,53]</td>
<td></td>
</tr>
<tr>
<td>mRNA</td>
<td>BAL cells</td>
<td>Higher [99]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>IL-10</td>
<td>Protein</td>
<td>ser, ebc</td>
<td>Higher [53,54]</td>
<td></td>
</tr>
<tr>
<td>Polymorph</td>
<td>Gene</td>
<td>Yes</td>
<td>Lower [55]</td>
<td></td>
</tr>
<tr>
<td>IL-11</td>
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<td>Gene</td>
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<td></td>
</tr>
<tr>
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<td>Protein</td>
<td>ser</td>
<td>Higher [54]</td>
<td></td>
</tr>
<tr>
<td>CCL2</td>
<td>mRNA</td>
<td>Lung tissue</td>
<td>Higher [30,101]</td>
<td></td>
</tr>
<tr>
<td>Protein</td>
<td>BAL</td>
<td>Higher [30]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CCR2</td>
<td>mRNA</td>
<td>Lung tissue</td>
<td>Higher [101]</td>
<td></td>
</tr>
<tr>
<td>Protein</td>
<td>Lung tissue</td>
<td>Higher [30]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CCL3</td>
<td>mRNA</td>
<td>Lung tissue</td>
<td>Higher [101]</td>
<td></td>
</tr>
<tr>
<td>Protein</td>
<td>Lung tissue</td>
<td>Higher [30]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CCL4</td>
<td>Protein</td>
<td>BAL</td>
<td>Higher [30]</td>
<td></td>
</tr>
<tr>
<td>CCR5</td>
<td>No. epi cells</td>
<td>Lung tissue</td>
<td>Higher [30]</td>
<td></td>
</tr>
<tr>
<td>CXCL1</td>
<td>Protein</td>
<td>sp, lung tissue</td>
<td>Higher [30]</td>
<td></td>
</tr>
<tr>
<td>CXCL8</td>
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<td>Higher [30,101]</td>
<td></td>
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<tr>
<td>Protein</td>
<td>ser, sp, BAL</td>
<td>Lung tissue</td>
<td>Higher [30]</td>
<td></td>
</tr>
<tr>
<td>CXCR2</td>
<td>Protein</td>
<td>PBL</td>
<td>Higher [30]</td>
<td></td>
</tr>
<tr>
<td>mRNA</td>
<td>BAL cells</td>
<td>Higher [99]</td>
<td></td>
<td></td>
</tr>
<tr>
<td>CXCL10</td>
<td>Protein</td>
<td>Lung tissue</td>
<td>Higher [17,30]</td>
<td></td>
</tr>
<tr>
<td>CXCR3</td>
<td>No. T+ cells</td>
<td>Lung tissue</td>
<td>Lower [17,30]</td>
<td></td>
</tr>
</tbody>
</table>

*polyorphism = polymorph; no. = number; epi = bronchial epithelial; ser = serum; sp = sputum; BAL = bronchoalveolar lavage; PBL = peripheral blood lymphocytes; ‘* = inverse correlation with FEV1; ‘† = especially in severe COPD; ‘expression or levels in COPD compared with controls; yes = polymorphisms reported.*
inflammation and the decline in lung function, and the side effects of steroids include increased risk of hip fractures, skin bruising and candidiasis [32]. Novel alternative inhibitors of inflammation include specific inhibitors of phosphodiesterase 4 (PDE4), leukotriene receptor antagonists, inhibitors of 5-lipoxygenase (leukotriene synthesis inhibitor) and cyclooxygenase (COX) inhibitors (that inhibit prostaglandin synthesis) [36]. These could be more promising as effective treatments for COPD. However, more-specific inflammatory therapies without adverse effects are needed. Antagonists for TNFα, IL-1, CXCL8 receptor CXCRI2, or CCL2/CCR2 and their receptors seem useful in Th1-type cytokine-driven diseases, such as rheumatoid arthritis (RA), psoriasis and inflammatory bowel disease or Crohn’s disease. This review focuses on the recent developments in this field.

**Potential cytokine drugs**

The general concept of cytokine therapy is given in Figure 3. Different intervention levels can be explored. The currently examined levels of intervention include: (1) scavenging and neutralization of cytokines by binding to soluble receptors or neutralizing antibodies; (2) inhibition of cytokine binding to its receptor by small compounds or incomplete and non-activating cytokines; (3) inhibition of protein activation (processing of preprotein into mature protein); and (4) inhibition of signal transduction and transcription (via inhibition of mitogen
activated protein kinase (MAPK) p38, nuclear factor κB (NFκB), inhibitors of NFκB (IKK and IκB), Cot kinase, and via antisense mRNA molecules for cytokines. Many potential drugs are still in preclinical development or in clinical trial phases for diseases other than COPD (Tables 2 and 3). A few are, however, in clinical trials, but no study data has yet been published for COPD in peer-reviewed journals. In the following sections, some potential drugs are highlighted.

**TNFα**

TNFα is a pro-inflammatory cytokine that activates and stimulates migration of monocytes, macrophages, neutrophils and T cells. In COPD, higher levels of this protein have been observed in induced sputum and plasma, whereas increased mRNA levels were found also in leg muscles (Table 1). Reports on gene-activating polymorphisms showed no unequivocal data but relied upon different populations (e.g. Japanese versus Caucasian) and polymorphisms (e.g. -376G/A; -308; -238G/A; +489G/A) [6,7,37]. However, the polymorphisms seem to be associated with a poorer prognosis [6,7]. TNFα was shown to play a role in cigarette smoke-induced emphysema and airway inflammation with neutrophils and macrophages in mice [38]. In addition, they also showed that following exposure to cigarette smoke, mice lacking TNFR p55 and p75 had threefold less increase in airspace enlargement, 65% fewer neutrophils and no increase in macrophage numbers in the bronchoalveolar lavage (BAL) fluid compared with wild-type mice [39]. MMP2 and MMP9 levels in BAL fluid were also not enhanced in these TNFR--/-- mice. These data point to TNFα as an attractive target for therapy of COPD.

Several drugs aimed at reducing TNFα levels have been developed for treatment of other chronic diseases such as (adult or juvenile) RA, psoriasis, and inflammatory bowel disease (especially Crohn’s disease). A few of these drugs (infliximab, etanercept and adalimumab) have been approved by the FDA and the EC for disease-specific application; whereas others (like afelimomab, CDP-870 and CDP-571) are being developed or in clinical trial for RA, ankylosing spondylitis (AS), psoriasis or Crohn’s disease (Tables 2 and 3). Of these drugs, only a few are in clinical trial (phase II) for asthma or COPD (Table 2). Activities on CDP-571 were reported to have been discontinued by its manufacturer, Celltech, because of market considerations.

TNFα antagonists include non-human or chimeric antibodies (infliximab, afelimomab and CytoTab), humanized antibodies (adalimumab, CDP-571, CDP-870), human TNFR (Onerecept) or TNFR fusion protein (Etanercept). The general route of application is subcutaneous in RA, AS, Crohn’s disease or COPD, and topically in psoriasis. Polyethylene glycolylation (Pegylation) of molecules (CDP-870; PEGylated TNFR1) protects TNFα-binding compounds from proteolytic breakdown, renders them potentially less immunogenic, and can improve bioavailability, half life and circulation time of poorly water-soluble drugs. Immunogenicity of therapeutic antibodies can be reduced, compared with corresponding murine molecules, by humanization. However, many clinical trials showed their effectiveness but also side effects. These adverse effects include reactions around the site of administration and, less frequently, a delayed hypersensitivity-like reaction, new onset of autoimmunity and on rare occasions, drug-induced systemic lupus erythematosus and demyelination, serious infections, vasculitis or malignancies can occur [40,41], which can lead to a loss of efficacy. To avoid the side effects that can arise from an invasive treatment strategy with frequent injections, smaller non-injectable, bioactive molecules are needed with long half-life and high bioavailability, capable of regulating TNFα expression,

**FIGURE 3**

A schematic diagram of the general pathway leading from gene expression to protein effect and potential points of intervention. Intervention levels are indicated: (1) regulation of gene expression by antagonists of MAPK p38, NFκB, IKK and Cot kinase or by cytokine-specific siRNA; (2) translation of mRNA into preprotein by antisense oligonucleotides; (3) protein processing by converting enzymes; (4) binding of the mature protein to its receptor by cytokine binding drugs, antibodies or receptor antagonists; and (5) by interference with the receptor signaling cascade using specific compounds including those mentioned above. Intervention might lead to inhibition of the effects shown in the lower boxes. ECM, extracellular matrix.
synthesis and/or secretion [42]. Among the small molecules capable of such effects are antisense oligonucleotides for TNFα (ISIS-104838), which is in clinical trial phase II for RA and Crohn’s disease (Table 2). This molecule inhibits TNFα mRNA translation into preTNFα proteins, finally resulting in reduced extracellular TNFα levels. ISIS-104838 shows a similar bioavailability for oral and intravenous (i.v.) administration [43]. ISIS Pharmaceuticals is now exploring the less invasive oral administration route as this is generally preferred to i.v. treatment. However, inhalation antisense formulations are less likely to reduce systemic pulmonary inflammation in COPD, but can be useful to treat local pulmonary inflammation.

With regard to asthma and COPD, there has been no follow-up to the small study by Babu et al. [44] treating severe asthmatics with etanercept. This study showed preliminary data on treatment with etanercept plus withdrawal of β2-agonists, resulting in improved FEV1 and stable FEV1/forced vital capacity. In vitro, a potential therapeutic effect of infliximab was described by inhibiting

### Table 2

Examples of anti-inflammatory cytokine antagonists in clinical trial potentially useful for treatment of COPD

<table>
<thead>
<tr>
<th>Name</th>
<th>Compound</th>
<th>Function</th>
<th>Trial phase (disease)</th>
<th>Company</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>TNFα</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Infliximab (Remicade)</td>
<td>Chimeric IgG1 human/mouse anti-TNFα antibody</td>
<td>Neutralization TNFα</td>
<td>FDA (RA, Crohn); III (AS, psoriasis); II (COPD)</td>
<td>Centocor/ Schering-Plough</td>
</tr>
<tr>
<td>Etanercept (Enbrel)</td>
<td>Human TNFR p75 :Fc IgG1 fusion protein</td>
<td>Neutralization TNFα</td>
<td>FDA (RA, AS); II (asthma)</td>
<td>Amgen/Wyeth</td>
</tr>
<tr>
<td>Adalimumab (Humira)</td>
<td>Human IgG1 anti-TNFα antibody</td>
<td>Neutralization TNFα</td>
<td>III (AS, Crohn) II (psoriasis)</td>
<td>Abbott</td>
</tr>
<tr>
<td>Afelimomab (Segard)</td>
<td>Mouse F(ab’)2 anti-TNFα fragment</td>
<td>Neutralization TNFα</td>
<td>III (sepsis)</td>
<td>Abbott</td>
</tr>
<tr>
<td>CDP-571 (Humidade)</td>
<td>Humanized mouse anti-TNFα antibody</td>
<td>Neutralization TNFα</td>
<td>III (Crohn)</td>
<td>Celltech/Biogen</td>
</tr>
<tr>
<td>CDP-870</td>
<td>Polyethylene-glycolated-human F(ab’) anti-TNFα fragment</td>
<td>Neutralization TNFα</td>
<td>III (RA, Crohn)</td>
<td>Celltech/Pfizer</td>
</tr>
<tr>
<td>Onercept</td>
<td>Rec. human sTNFR p55</td>
<td>Neutralization TNFα</td>
<td>II (Crohn, psoriasis)</td>
<td>Serono</td>
</tr>
<tr>
<td>PEG sTNF-R1</td>
<td>Polyethylene-glycolated-human rec. soluble TNF receptor p55</td>
<td>Neutralization TNFα</td>
<td>II (RA)</td>
<td>Amgen</td>
</tr>
<tr>
<td>CytoTab</td>
<td>Sheep F(ab’) anti-TNFα antibody</td>
<td>Neutralization TNFα</td>
<td>Iib (sepsis); I (Crohn)</td>
<td>Protherics</td>
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<tr>
<td>ISIS-104838</td>
<td>Antisense TNFα methoxy-oligonucleotide</td>
<td>Inhibits TNFα transcription</td>
<td>II (RA, Crohn)</td>
<td>ISIS/Elan</td>
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<td><strong>IL-1/IL-10</strong></td>
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<td>Anakinra (Kineret)</td>
<td>Rec. human IL-1Ra IL-1</td>
<td>Neutralization</td>
<td>FDA (RA) ; II (Crohn)</td>
<td>Amgen</td>
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<td>Human IL-1–Fc IgG1 fusion protein</td>
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<td>Regeneron</td>
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<td>CDP 484</td>
<td>PEGylated F(ab’) fragment against IL-1β</td>
<td>Neutralization IL-1β</td>
<td>I (RA)</td>
<td>Celltech</td>
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<td>Ilodecakin (Tenovil)</td>
<td>Recombinant human IL-10</td>
<td>Agonist for IL-10R</td>
<td>III (Crohn) ; II/III (RA, psoriasis)</td>
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<td><strong>CCL2/CCR2</strong></td>
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<td>MLN-1202</td>
<td>Humanized mouse anti-CCR2 antibody</td>
<td>Inhibits binding</td>
<td>II (RA)</td>
<td>Millennium</td>
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<td>INC83284</td>
<td>CCR2 antagonist</td>
<td>Inhibits binding CCL2 to CCR2</td>
<td>I (DTH, RA)</td>
<td>Incyte</td>
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<tr>
<td><strong>CCR5</strong></td>
<td></td>
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</tr>
<tr>
<td>AK602 (ONO4128; GW873140)</td>
<td>Spirodiketopiperazine derivative</td>
<td>Inhibits CCL3 binding to CCR5</td>
<td>I (HIV)</td>
<td>Kumamoto University (JP)</td>
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<tr>
<td><strong>CXCR2</strong></td>
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<tr>
<td>SB332235</td>
<td>CXCR2 antagonist</td>
<td>Inhibits binding CXCL8 to CXCR2</td>
<td>I (COPD)</td>
<td>GlaxoSmithKline</td>
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<td>SB656933</td>
<td>CXCR2 antagonist</td>
<td>Inhibits binding CXCL1 to CXCR2</td>
<td>I (COPD)</td>
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</tr>
<tr>
<td><strong>Others</strong></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Pirfenidone (Deskar)</td>
<td>Pyridine derivate</td>
<td>Inhibits cytokine production via NFκB</td>
<td>I (MS)</td>
<td>Shionogi</td>
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<tr>
<td>BMS-561392</td>
<td>TACE and MMP Inhibitor</td>
<td>Blocks pre-TNFα processing</td>
<td>II (RA, Crohn)</td>
<td>BristolMyersSquibb</td>
</tr>
</tbody>
</table>

Abbreviations: AS, ankylosing spondylitis; DTH, delayed type hypersensitivity; HIV, human immunodeficiency virus; MS, multiple sclerosis; RA, rheumatoid arthritis; FDA, approved by the US federal food and drug administration.
the TNFα-induced expression of mucin SAC (MUC5AC) by an airway epithelial cell line [45]. Currently, we are awaiting data from ongoing clinical trials to discuss efficacy and side effects.

**TACE inhibitors**

Experimental TNFα antagonists include the inhibitors of TNFα converting enzyme (TACE; Table 3). These molecules inhibit pre-TNFα cleavage into mature TNFα and are inhibitors of MMPs. Recent studies demonstrated that cigarette smoke and lipopolysaccharide-(LPS) induced mucin 5AC secretion from human lung epithelial cells [46]. As mucins 5AC are important constituents of mucus seen in chronic bronchitis, inhibition of TACE via TACE-specific drugs would appear to be potentially beneficial. Of this class of potential drugs, two are in clinical trial. Marimastat is in clinical trial for pancreatic and gastric cancer as an MMP inhibitor while BMS-561392 (or: DPC-333) is in evaluation as a TACE inhibitor in RA. However, side effects of Marimastat include musculoskeletal pain, inflammatory polyarthritis and hand contraction, which could preclude its applicability. The concomitant inhibition of MMP activity might be important in reestablishment of the protease–antiprotease balance in COPD, which favours proteolytic breakdown of tissue at higher MMP and elastase activities. Several other novel TACE compounds are still in preclinical development stage and have to prove their effectiveness [30]. Among these is TMI-1, a thiomorpholinecarboxamide derivative, which specifically inhibits LPS-induced TNFα secretion but not secretion of other proinflammatory cytokines such as IL-1β, IL-6 or IL-8, both from human cell lines and in human explants [47]. In vivo, TMI-1 reduced clinical symptoms in a collagen-induced arthritis model in mice [47]. Furthermore, TACE-specific, small interfering RNA (siRNA) which blocks TACE mRNA synthesis, might prove to be useful, but little is known about the clinical effects yet.

In conclusion, novel TNFα antagonists are still being developed, whereas older ones have proven to be selectively effective in treatment of specific diseases. However, data on the treatment of patients with asthma or COPD are still not available in peer review journals, precluding discussion on efficacy and side effects in these diseases. Given the selective efficacy of TNFα antagonists, clinical trials for COPD with different potential antagonists seems worthwhile for determining efficacy.

**Interleukins**

The interleukins form a large family of proteins with diverse immunological effects. Pro-inflammatory interleukins include IL-1 and IL-6, whereas interleukins such as IL-10 and IL-11 are anti-inflammatory cytokines. IL-10 stimulates T helper null (Th0) cells to differentiate into Th2 cells that express cytokines like IL-4 and IL-5, and hence drives Th2-like reactions seen in, for example, allergic diseases. It inhibits transcription and translation of Th1-like cytokines (IL-6, TNFα), and reduces natural

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**Table 3**

<table>
<thead>
<tr>
<th>Name</th>
<th>Compound</th>
<th>Function</th>
<th>Trial phase (disease)</th>
<th>Company</th>
</tr>
</thead>
<tbody>
<tr>
<td>CCL2/CCR2</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>RS-504393</td>
<td>Spiropiperidine</td>
<td>Inhibits CCL2 binding to CCR2</td>
<td>Preclinical</td>
<td>Roche</td>
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<tr>
<td>MCP-1α</td>
<td>CCL2 peptide equal to amino acids 9 to 76</td>
<td>Inhibits CCL2 binding to CCR2</td>
<td>Preclinical</td>
<td>University of British Columbia</td>
</tr>
<tr>
<td>CCR5</td>
<td>TAK220</td>
<td>Inhibits CCL3 binding to CCR5</td>
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<td>CXCR2</td>
<td>SB265610</td>
<td>Inhibits CXCL1/8 to CXCR2</td>
<td>Preclinical</td>
<td>GlaxoSmithKline</td>
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<td></td>
<td>SB225002</td>
<td>Inhibits CXCL1/8 to CXCR2</td>
<td>Preclinical</td>
<td>GlaxoSmithKline</td>
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<tr>
<td></td>
<td>GROαβ8</td>
<td>Inhibits CXCL1 binding to CXCR2</td>
<td>Preclinical</td>
<td>University of Bern</td>
</tr>
<tr>
<td>Others</td>
<td>Bindarit</td>
<td>Inhibits TNFα and CCL2 synthesis</td>
<td>Preclinical</td>
<td>Angelini Pharmaceutical</td>
</tr>
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<td></td>
<td>Antileukinate</td>
<td>Antagonist CXCR</td>
<td>Preclinical</td>
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<td></td>
<td>NR-58–3.14.3</td>
<td>Chemokine-induced migration antagonist</td>
<td>Preclinical</td>
<td>University of Cambridge (UK)</td>
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<td></td>
<td>Pioglitazone (Actos/Glitazin)</td>
<td>Reduction CCR2 expression; PPARγ activation</td>
<td>Preclinical (FDA as PPARγ agonist in DM2)</td>
<td>Takeda/Eli Lilly</td>
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<td></td>
<td>GW3333</td>
<td>TACE and MMP Inhibitor</td>
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<td>TACE and MMP Inhibitor</td>
<td>III (cancer) as MMP inhibitor</td>
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</table>

Abbreviations: DM2, diabetes mellitus type 2; PPARγ, peroxisome proliferator-activated receptor γ
killer cell and macrophage activity. Macrophages, Th2 cells and CD8+ T cells produce IL-10. IL-12 derived from dendritic cells (DCs) stimulates differentiation of Th1 cells associated with interferon γ synthesis, pointing to the involvement of IL-12 in Th1-like reactions. Hence, IL-10 and IL-12 balance the Th1 and Th2 reactions. IL-1 is produced by macrophages or epithelial cells upon exposure to stimuli like cigarette smoke or lipopolysaccharide (LPS). It stimulates TNFα expression in macrophages. IL-1 has two naturally occurring soluble antagonists, sIL-1Ra and sIL-1RII, that compete for binding of IL-1 to its receptors. Many cell types, including epithelial and endothelial cells, fibroblasts, macrophages and T cells produce IL-6. It is generally expressed during inflammation or tissue damage and can be induced by cytokines like IL-1 or TNFα. IL-6 has pleiotropic effects including generation of cytotoxic T cells. Its natural antagonist is the soluble IL-6 receptor (gp80, or CD126) which is present in blood.

With respect to COPD, a recent biopsy study showed that COPD patients have more bronchial T cells with activated Th1-type associated transcription factor STAT4 linked with a higher number of IFNγ+ cells compared with subjects without COPD [48]. This supports the idea that COPD is a Th1-mediated disease. One study reported enhanced levels of IL-1 in induced sputum from patients with mild COPD as compared to healthy subjects [49]. Other studies did not find any difference in IL-1 levels either in serum, sputum or BAL fluid. One study reported enhanced serum levels of its antagonist sIL-1Ra in COPD [50], whereas sIL-RII serum levels did not differ from that in controls without COPD [51]. For IL-6 or sIL-6R, two reports mention higher IL-6 protein levels in exhaled breath condensate [52,53], but altered levels have not been found in other body compartments in COPD patients. With respect to IL-10, protein levels can either be higher or lower in COPD depending on the compartment examined. Higher levels were observed in serum and exhaled breath condensate of COPD patients [53,54], but lower levels in sputum [55] compared with non-smoking controls or smokers without COPD. In addition, several studies pointed to transcription altering point mutations in the promoter region of the IL-10 gene associated with COPD [56–58]. However, each study found a different site to be associated with COPD, which might be dependent on the examined population, COPD subgroup, or promoter site. IL-12 protein levels were found, only in one study, to be higher in COPD in serum [54]. Despite these differences, which can reflect differential expression in various body compartments, these data suggest the involvement of IL-1 and IL-10 in the pathogenesis of COPD. The outcome of clinical trial studies on other Th1-type diseases may provide evidence to start interleukin antagonist studies for COPD.

**IL-1 antagonists**

Reduction of IL-1 levels can be achieved via the sIL-1Ra, using IL-1 binding proteins like IL-1 Trap, IL-1β or IL-1 receptor antibodies, and inhibitors of IL-1β converting enzyme (ICE). Anakinra, a recombinant IL1 receptor antagonist, has been approved for treating RA. Other IL-1 receptor antagonists, IL-1 Trap and CDP484 are currently in clinical trials (Table 2). Both Anakinra and IL-1 Trap bind and neutralise circulating IL-1 via their soluble IL-1 receptor moiety, whereas CDP 484 is an anti-IL-1β antibody. These drugs are administered via subcutaneous injection. Clinical studies on anakinra demonstrated a reduction in the pathology and symptoms of RA [59]. Frequent side effects include mild reactions at injection site and an increased risk of infection. In patients with previous pneumonia or COPD, administration of anakinra increased the risk of bacterial lung infection. Clinical data on IL-1 Trap and CDP484 are, as yet, limited [30]. A recent *in vitro* study by Werman et al. [60] pointed to intracellular IL-1α preprotein being an activator of transcription factors NFκB and AP-1. Under conditions of endotoxin activation or IL-1 receptor blockade, cells overexpressing IL-1α showed nuclear translocation, activation of NFκB and AP-1, and higher expression of CXCL8 and IL-6. Hence, extracellular reduction of IL-1 might not be sufficient to reduce inflammation. Despite this, the use of IL-1 antagonists in COPD could potentially lower IL-1 levels to reduce proinflammatory cytokine expression like TNFα.

**IL-10 agonist**

Only one drug of the IL-10 agonist class (ilodecakin) is currently in clinical trial (Table 2). This agonist is recombinant human IL-10 and is being tested for efficacy in various diseases including psoriasis, Crohn’s disease, RA, and Hepatitis C [61]. Ilodecakin is administered subcutaneously, either daily or less frequently. Pretreatment with IL-10 reduced the expression of cytokines like TNFα, IL-6, CXCL8, CCL2, CCL3 and CCL4 as well as pulmonary neutrophilia in healthy volunteers following exposure to endotoxin [62]. *In vitro*, IL-10 inhibits TNFα and CXCL8 production by macrophages, but induces tissue inhibitor of metalloprotease (TIMP-) 1 and does not affect MMP9 expression [29,61]. One of the mechanisms of IL-10 action is through the blockade of IκB Kinase (IKK) (which consequently impairs NFκB function) and via selective stimulation of NFκB p50/p50 translocation into the nucleus [64]. This homodimer of NFκB represses inflammatory gene transcription. As in COPD, sputum IL-10 levels are reduced, TNFα and CXCL8 levels are elevated and the protease-antiprotease balance is shifted to tissue degradation, these data suggest that IL-10 treatment of COPD patients might be beneficial.

However, clinical studies on other diseases somewhat temper this hypothesis. Clinical studies on psoriasis and psoriatic arthritis using subcutaneous IL-10 injections, generally produced clinical improvement in skin disease, accompanied by decreased expression of Th1 cytokines [61]. It is hypothesized that this is due to suppression of DC maturation and macrophage activities, reduced TNFα
and IL-12 expression and shifting from Th1 to Th2 type T cells. Several studies demonstrated that treatment for up to one year with IL-10 (daily or three times a week) of patients with chronic hepatitis C infection and liver fibrosis, resulted in a shift from IFNγ+CD8+ T cells into a Th2-type profile, improved liver histology and reduced fibrosis but increased viral loads [61,65]. By contrast, the limited number of studies with IL-10 treatment in RA showed a rather small clinical improvement [61]. Also, in Crohn’s disease, IL-10 treatment did not result in marked clinical improvement. This may be due to IL-10-induced IFNγ expression and IFNγ-dependent elevation of CXCL10 expression, which in turn may result in counterbalancing proinflammatory reactions [61]. This effect was also shown in a mouse tumour model. Treatment of mice with recombinant IL-10 resulted in tumour growth inhibition and enhanced levels of CXCL9 and CXCL10. Treatment with neutralising antibodies against CXCL9 or CXCL10 of mice bearing tumours overexpressing IL-10 resulted in reduced tumour growth inhibition, suggesting an interaction between IL-10 and CXCL10 activities [66]. Hence, IL-10 treatment of patients may result in higher CXCL10 levels and subsequent increased inflammation with CXCR3+ T cells as seen in COPD. One recent study noted that treating COPD patients for 3 months with the corticosteroid VentolaR, caused enhanced release of IL-10 levels from alveolar macrophages ex vivo [67]. However, the FEV\textsubscript{1} was not improved by this treatment compared with placebo treated subjects. No data on airway inflammation were reported. This suggests that therapies for COPD that lead to augmented IL-10 levels might not result in an improvement in FEV\textsubscript{1}.

The adverse effects of recombinant IL-10 treatment include anaemia and headache. In addition, the recombinant protein may elicit immunological reactions. Hence, smaller molecules may be useful, but may easily be scavenged by binding proteins. Given these data, further investigations into the role of IL-10 in the pathogenesis of COPD should be performed before clinical trials with IL-10 with COPD patients start.

In conclusion, treatment with IL-1 antagonists or IL-10 showed improvements in clinical symptoms and were capable of reducing proinflammatory cytokine expression in psoriasis. Whereas IL-1 antagonist therapy in COPD might be effective in reducing inflammation, the lack of a pathogenetic basis for IL-10 therapy linked with the potential for adverse effects, presently precludes IL-10 therapy. In view of this, the therapeutic effects of the IL1Ra–IL-10 fusion protein (recently reported to reduce immunopathology in a rat arthritis model) cannot be considered yet for models of COPD.

**Chemokines**

Airway inflammation in COPD has been reported to consist mainly of CD68+ macrophages, elastase-positive neutrophils and CD8+ T cells. Physiologically, each of these cells is activated by a specific subset of chemokines. Macrophages migrate towards a gradient of CCL2, CCL3 or CCL4 via their receptors CCR2 or CCR5. Maus et al. [68] recently demonstrated, using CCR2 deficient mice, that alveolar monocyte migration \textit{in vivo} is only dependent on CCR2 activation. Neutrophils are mainly attracted by CXCL1 and CXCL8 via CXCR1 and CXCR2, whereas CXCR1 has a larger affinity for CXCL8 than for CXCL1. CXCR1 is involved in neutrophilic degranulation, protease release and oxidative stress. T cells can have receptors for CCL2 to CCL4, and a subset also has CXCL10 receptors (CXCR3).

In COPD, higher levels of these chemokines and their receptors have been reported either to be (inversely) associated with lung function or not to be associated (Table 1). Several studies using \textit{in vitro} models with cigarette smoke condensate exposition or \textit{in vivo} models for COPD showed a similar inflammatory cell pattern and chemokine and cytokine expression pattern [17,21,30]. Hence, these chemokines are thought to be involved in the pathogenesis of COPD. Therefore, it has been proposed that chemokine antagonists might have potential as specific treatments for inflammation in COPD. Many chemokine and chemokine receptor antagonists are being developed [30], but only a few are in clinical trials for inflammatory diseases (Table 2). Table 3 refers to recently (further) developed compounds.

**CCL2/CCR2 antagonists**

Of the CCL2 antagonists, the humanised mouse monoclonal anti-CCL2 antibody MLN1202 is in phase II for treatment of RA. According to Millennium, a single dose phase I trial study did not reveal serious adverse effects and it was well tolerated by healthy volunteers. According to Incyte, the selective CCR2 antagonist INCB3284 binds to CCR2 but not CCR1 or CCR5, and prevents CCL2 from binding to CCR2. This drug has now entered phase I trials for single and multiple dose testing in healthy volunteers that will also investigate delayed type hypersensitivity. Future phase II studies with INCB3284 will be directed to treatment of RA. The main advantage of this drug over MLN1202 is that it can be administered by the oral route. As presented at the 3rd Annual Needham Biotechnology Conference (New York, 2004), other CCR2 antagonists by Incyte are being developed. A recent study on RS-504393 indicated that this compound blocked macrophage infiltration, activation, CCL2 expression and CCR2-dependent renal fibrosis in mice [69]. At this stage, the clinical applicability and potential adverse adrenergic effects of this drug have not yet been established [30]. Interestingly, a set of eosinophil-chemotactic chemokines (CCL11/eotaxin-1 and CCL26/eotaxin-3) that bind to CCR3, have been shown to antagonize CCL2-dependent signalling of CCR2 and CCR5, thereby inhibiting chemotaxis and actin polymerisation of monocytes [70,71]. This suggests a potential antagonistic effect on monocyte/macrophage inflammation in COPD patients without eosinophilia via the natural ligand CCL11 or CCL26.
Another CCL2 antagonist is bindarit, a benzyl-indazolic compound, which has been shown to inhibit CCL2 and TNFα but not CXCL8 expression by monocytes in vitro. It was shown to reduce monocyte and leukocyte numbers in addition to CCL2 expression in mice treated with carrageenan and in a mouse model for lupus arthritis [72]. In an experimental mouse model of arthritis, bindarit administered orally reduced clinical symptoms as well as CCL2 and TNFα but not IL-1 or IL-6 expression [73]. As both CCL2 and TNFα expression were enhanced in COPD in addition to higher numbers of macrophages and CD8+ T cells, bindarit or compounds with similar antagonistic features may prove to be useful for the treatment of inflammation in COPD. However, no data related to safety and tolerability are currently available.

Macrophages can also migrate along gradients of CCL3 or CCL4 via their receptor CCR5. As the expression of both chemokines was found to be increased in COPD, treatment with antagonists might be useful. In this respect, two antagonists can be mentioned, TAK220 and AK602 (Tables 2 and 3). These drugs are presently being investigated for the treatment of human immunodeficiency virus infections of CCR5+ cells (T cells and macrophages), but also have the capacity of inhibiting binding of CCL3 to CCR5. However, as CCL4 binding is not inhibited by these compounds, activation or chemoattraction of macrophages in COPD has not been clarified, further studies on their role in COPD will be necessary before CXCL10 antagonist-based therapy can be established.

**CXCL and CXCR antagonists**

As CXC chemokines seem to be involved in the pathogenesis of COPD, many antagonistic compounds have been developed that showed inhibitory effects on binding of CXCL1 and CXCL8 to their receptor CXCR2 in vitro [30]. One compound is now being evaluated in clinical trial phase I for COPD (SB332235) (Table 2). SB332235 is a non-peptide inhibitor of CXCL1 binding to CXCR2, which impairs CXCL1 and CXCL8-induced chemotaxis of neutrophils, T cells, and DCs. However, human safety and pharmacokinetic data have not been published yet. The oral CXCR2 antagonist SB656933 presented at the 14th European Respiratory Society Congress (Glasgow, 2004) was reported to inhibit CXCL1- and CXCL8-stimulated CXCR2 but not CXCR1 activation in human neutrophils. It could also reduce CXCL1-induced neutrophil CD11b expression both in vitro and in vivo, and it impaired recruitment of CD11b+ neutrophils in the BAL fluid from LPS aerosol-exposed rats [74,75]. This compound now entered clinical phase I studies for COPD.

The first data from studies on treatment of COPD patients with an antibody against CXCL8 (ABX-IL-8) were recently presented by Mahler et al. [76]. In this clinical phase II study, COPD patients were treated with three consecutive injections (800mg at baseline and 400mg in the two subsequent months). No significant adverse effects were noted upon ABX-IL-8 treatment. The severity of dyspnea was reduced whereas no improvements were observed in lung function (FEV1, FVC, TLC), quality of life or the six-minute walking test. Although effects on pulmonary inflammation, the efficacy of different doses and long term treatment effects as well as effects upon stratification of patients according to their GOLD stage were not determined, this study points to potential treatment effects of this (type of) antagonist. However, Abgenix reported in 2003 that ABX-IL-8 will be withdrawn from further studies because the effects achieved in treatment of RA were too small and because of market considerations.

Two other compounds, SB225002 and SB265610, are different biphenyl urea derivatives that inhibit binding of CXCL1 and CXCL8 to CXCR2 by interfering with the chemokine-binding sites [30]. Of these, the oral efficacy of SB265610 treatment was established by the inhibition of repair and neutrophil recruitment in a skin-wound healing model in mice [77]. In addition, two other novel compounds have been described. Widdowson et al. [78] reported a highly selective N,N-diarylurea CXCR2 antagonist, capable of reducing neutrophil chemotaxis in vitro, and inhibiting neutrophilia in rabbit models for ear swelling and neutropenia. Baxter et al. [79] reported a new triazolothiel that was an effective oral CXCR2 antagonist.

CXCL10 is a chemokine secreted by cells stimulated by IFNγ or LPS as seen in many Th1-type inflammatory diseases. In addition to its role in inflammation, CXCL10 stimulates smooth muscle cell proliferation and migration in vitro, and inhibits angiogenesis in vivo in mice by inhibition of endothelial cell differentiation [80]. At present, antagonists for CXCL10 or CXCR3, have only been used in the laboratory and the status of preclinical development is not clear. The effectiveness of a CXCL10 antibody in vivo was shown in a mouse model for T cell-associated disease showed demonstrating impaired CXCR3+ T cell recruitment [81]. As the role of CXCR3+ T cells in COPD has not been clarified, further studies on their role in COPD will be necessary before CXCL10 antagonist-based therapy can be established.

**Broad-spectrum chemokine antagonists**

Drugs that can inhibit the expression of multiple cytokines or chemokines could represent an alternative strategy. Pirfenidone, an antifibrotic pyridine derivative, was shown to inhibit the expression of TNFα, IFNγ, IL-6 and IL-12, whereas it induced the expression of IL-10 in mouse models of endotoxic shock [82–84]. In these models, pirfenidone, given either before or after LPS treatment, inhibited lethal shock and mortality dose-dependently. Mice pretreated with pirfenidone showed reduced airway neutrophilia and TNFα levels compared with controls, following aerosol challenge with LPS [82]. The mechanism was reported to be via inhibition of NFκB [84]. Because of its anti-inflammatory effects, pirfenidone is now in clinical trial phase I for multiple sclerosis, but could be useful for COPD as well.
Another broad-spectrum chemokine antagonist is NR58–3.14.3, which is a retroinverso analogue of the mature CCL2 amino acids 51–62 [85]. This antagonist was shown to inhibit leukocyte migration to CCL2, CCL3, CCL5, CXCL8 and CXCL12 in vitro [85]. In vivo, NR58–3.14.3 inhibited leukocyte migration and TNFα expression following intratracheal injection of LPS in rat skin [85]. In models of pulmonary ischemia-reperfusion, rats showed increased TNFα protein levels and neutrophil numbers in the BAL fluid, vascular injury, and lung TNFα mRNA levels, which could be reduced by up to 35% by pretreatment of the animals with NR58–3.14.3 [86]. These data show the chemokine and cytokine inhibitory function of and associated improvement in pathology caused by NR58–3.14.3. As NR58–3.14.3 is a small peptide analogue, it might prove to be less immunogenic and more successful as a therapy.

In conclusion, several single-chemokine antagonists or receptor inhibitors are available. A few are in clinical trial and two of these specifically for the treatment of COPD. Several studies support the idea that monotherapy is less effective than multiple treatment in Th1-type inflammatory disease. A recent in vivo study showed that neutrophilic lung inflammation upon CCL2 or LPS instillation depends on CCR2+ monocytes [68]. In addition, in a mouse model of arthritis, inhibition of inflammation as well as improvement in clinical symptoms and pathology was achieved using a combination of CCL2 and CXCL1 inhibitors. The combination effect was shown to be greater than for the CCL2 inhibitor MCP-1 alone [87]. Finally, a study on peripheral blood monocytes from non-smokers and smokers with or without COPD reported that monocytes from COPD patients showed higher chemotactic responses to CXCL1 but not to CXCL8 or CCL2 compared with other subjects [88]. This suggests that single-chemokine targeting antagonists will probably affect either macrophage or neutrophil driven inflammation, whereas broad spectrum cytokine antagonists or combined treatment with CCL2/CCR2 and CXCL8/CXCR2 antagonists might be more effective in treating inflammation in COPD. However, a potential drawback could be the expression of functional chemokine receptors CCR2 and CXCR2 on lung epithelial cells that are probably involved in epithelial migration and wound healing [21,89]. Inhibition of these receptors might impair wound healing following smoke exposure and hence, could increase risk of infection and concomitant inflammation.

Transcription and signal transduction
An alternative route either to reduce cytokine or chemokine levels or to impair their effects is to interfere with gene expression or intracellular signal transduction. For both of these, signal transduction molecules like Nuclear Factor κB (NFκB) and Mitogen Activated Protein Kinase (MAPK) p38 are needed. NFκB is both a signal transduction and a transcription factor. In its inactive state it is bound to another protein, Inhibitor of κB (IκB). Upon phosphorylation of IκB by IκB Kinase (IKK), IκB dissociates from the complex and becomes ubiquitinated and subsequently degraded. Then, activated NFκB is translocated to the nucleus and binds to NFκB binding sites in the promoter region of genes such as cytokines and chemokines. MAPK p38 is activated via upstream signalling proteins including G-protein coupled receptors. MAPK p38 is involved in the regulation of transcription factors and a variety of enzymes including those involved in histone remodelling. Another class of kinases regulate activation of signalling proteins. Cot kinase, or Tpl 2, is a serine/threonine MAPK kinase kinase involved in activation of the kinase cascade leading to activation of e.g. MAPK p38, IKK and Jun Kinase (JNK) and subsequent transcription activation of genes like those for TNFα, IL-1 and CXCL8. Cot kinase is activated upon activation of TNFR p55 or the LPS receptor complex CD14 -Toll Like Receptor (TLR) 4.

Several studies have suggested that NFκB and MAPK p38 are involved in the pathogenesis of COPD [90,91]. The therapeutic effects of antagonists might be achieved at the transcription level of these signalling molecules, by blockade of intracellular translocation of NFκB, reduction of signalling of NFκB by inhibition of IκB degradation and inhibition of enzyme activity [92]. Many antagonists or inhibitors of MAPK p38, NFκB, IKK or Cot kinase are already in clinical trial phase II or II. Of the MAPK p38 antagonists, RWJ67657 (Johnson and Johnson) is in clinical trial phase I and has been shown to reduce cytokine expression levels and endotoxin-induced effects. The drug is indicated for use in RA, Crohn’s, psoriasis and allergic rhinitis.

SCIO-469 (Scios) is an oral antagonist of MAPK p38 and in clinical trial phase II for RA and multiple myeloma. Several studies presented at the annual conference of the American Thoracic Society (Orlando, USA, 2004) showed the effectiveness of p38 inhibitors in airway macrophages. Of the MAPK p38 inhibitors, sp-282 reduced the LPS-induced expression of TNFα by human lung macrophages in vitro by 70%, without affecting the expression of CXCL8 [93]. Pretreatment of human alveolar macrophages in vitro with BIRB796 and SB239063 demonstrated that they were more potent inhibitors of LPS-induced TNFα release than RO3201195 [94]. As MAPK p38 has four isozymes and each inhibitor has its own specificity towards one or more of these isozymes, this specificity could explain the experimental differences.

Recently, IKK antagonists including BMS345541 (Bristol-Myers Squibb) and SPC600839 (Celgene/Serono) have been developed, both of which are orally available, low-molecular weight inhibitors of IKK capable of reducing arthritis in murine models. BMS345541 was shown to impair NF-kB nuclear translocation and TNFα and IL-1β expression, and ameliorated the clinical and pathological symptoms of collagen-induced arthritis in mice in a...
dose-dependent manner [95]. Studies on pulmonary inflammation in COPD have, however, yet to be performed. With respect to NFkB, a recent study reported on the potential therapeutic effect of an siRNA directed against the NFkB subunit p65 in airway epithelial cell lines [96]. In this in vitro study, cells pretreated with this siRNA showed an impaired TNFa -induced NFkB p65, IL-6 and CXCL8 expression. An antisense antagonist for NFkB p65 is now in clinical trial phase I for Crohn’s disease [97].

In conclusion, although MAPK p38 and NFkB antagonists are at a more advanced stage with respect to clinical trials, antagonists for IKK and Cot kinase might also prove to be useful anti-inflammatory therapies. The potential disadvantage of using inhibitors of cell signalling molecules is that MAPK p38, NFkB, IKK and Cot kinase are general signal transduction proteins involved in various physiological processes. Systemic administration of antagonists can have profound effects on other receptor-mediated pathways other than the pro-inflammatory reactions in COPD. Moreover, inhibition of these molecules might also potentially impair defence mechanisms, particularly with respect to fighting infection.

Perspectives and concluding remarks
Cytokine antagonist drugs, capable of reducing TNFa expression or protein levels, are available and seem to be effective in different inflammatory diseases. Also, chemokine antagonists against the CCL2-CCR2 axis or CXCL1/CXCL8-CXCR2 axis are being tested now and might inhibit more specific parts of the inflammatory network. Addressing neutrophil- and macrophage-driven inflammation in COPD by the use of combined specific antagonists or broad-spectrum antagonists, including those impairing signal transduction routes, could also prove to be an interesting strategy. Although treatment with IL-1 antagonists or IL-10 is in an earlier stage of development and will probably require more pathophysiological insight into the function of these proteins in COPD, it might also represent a useful alternative approach. A complete block of cytokine-driven mechanism could be disadvantageous as defense reactions to infection can be impaired. In the short term, elevated levels of bacterial infection can be overcome by the use of antibiotics. This of course is of no use as a strategy for the treatment of opportunistic viral infection.

As COPD is more prevalent in the elderly, the long-term use of antagonists will further repress the already impaired immune system and potentially increase mortality due to infections. However, the efficacy and bioavailability of biologic drugs can be improved by reducing immunogenicity and improving routes of administration. Refining the end points for establishing drug efficacy in COPD will be of significant help in clinical assessment. Currently one of the major end points is the FEV1, which reflects the severity, but not the systemic effects of COPD. Although restoring the FEV1 is important, inclusion of other composite clinical and physiological parameters, as given in the BODE index for example, will provide additional functional information to assess efficacy [98]. In addition, as smoking is the main cause for COPD, stopping smoking should accompany any anti-cytokine therapies. Because quitting smoking alone is not enough to reduce pulmonary inflammation, it would be hoped that the combination should do so. Finally, restoration of lung tissue structure might be provided by other drugs like retinoic acids. Such approaches, however, will have to wait for the first results of cytokine antagonist therapy in COPD.

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