Cytokine modulators as novel therapies for airway disease

P.J. Barnes

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ABSTRACT: Cytokines play a critical role in orchestrating and perpetuating inflammation in asthma and chronic obstructive pulmonary disease (COPD), and several specific cytokine and chemokine inhibitors are now in development for the future therapy of these diseases.

Anti-interleukin (IL)-5 is very effective at reducing peripheral blood and airway eosinophil numbers, but does not appear to be effective against symptomatic asthma. Inhibition of IL-4 with soluble IL-4 receptors has shown promising early results in asthma. Inhibitory cytokines, such as IL-10, interferons and IL-12 are less promising, as systemic delivery causes side-effects. Inhibition of tumour necrosis factor-α may be useful in severe asthma and for treating severe COPD with systemic features.

Many chemokines are involved in the inflammatory response of asthma and COPD and several low-molecular-weight inhibitors of chemokine receptors are in development. CCR3 antagonists (which block eosinophil chemotaxis) and CXCR2 antagonists (which block neutrophil and monocyte chemotaxis) are in clinical development for the treatment of asthma and COPD respectively.

Because so many cytokines are involved in asthma, drugs that inhibit the synthesis of multiple cytokines may prove to be more useful; several such classes of drug are now in clinical development and any risk of side-effects with these nonspecific inhibitors may be reduced by the use of inhalational route of delivery.
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Correspondence: P.J. Barnes
Dept of Thoracic Medicine
National Heart & Lung Institute
Imperial College
Dovehouse Street
London SW3 6LY
UK
Fax: 0207 3515675

Cytokines play a critical role in the orchestration of chronic inflammation in all diseases, including asthma and chronic obstructive pulmonary disease (COPD). Multiple cytokines and chemokines have been implicated in the pathophysiology of asthma [1, 2]. There is less understanding of the inflammatory mediators involved in COPD, but, as this inflammatory process is markedly different from that in asthma, it is probable that different cytokines and chemokines are involved and that therapeutic strategies may therefore have to differ [3]. There is currently an intensive search for more specific therapies in asthma and for any novel therapies that may prevent the progression of airflow limitation in COPD. Inhibitors of cytokines and chemokines figure prominently in these novel therapeutic approaches [4, 5] (table 1).

Strategies for inhibiting cytokines

There are a number of possible approaches to the inhibition specific cytokines [6, 7]. These include drugs that inhibit cytokine synthesis (glucocorticoids, ciclosporinA, tacrolimus, myophenolate-helper lymphocyte (Th2)-selective inhibitors), humanized blocking antibodies to cytokines or their receptors, soluble receptors that mop up secreted cytokines, low-molecular-weight receptor antagonists and drugs that block the signal transduction pathways activated by

Table 1. – Potential cytokine modulators for asthma and chronic obstructive pulmonary disease therapy

Anticytokines	Inhibitory cytokines	Chemokine inhibitors	Cytokine synthesis inhibitors
Anti-IL-5	IL-1 receptor antagonist	CCR3 antagonists	Corticosteroids
Anti-IL-4	IL-10	CCR2 antagonists	Immunomodulators
Anti-IL-13	IL-12	CCR4 antagonists	Phosphodiesterase 4 inhibitors
Anti-IL-9	Interferons	CXCR2 antagonists	NF-κB inhibitors
Anti-IL-1	IL-18		p38 MAP kinase inhibitors
Anti-TNF-α			

IL: interleukin; TNF-α: tumour necrosis factor-α; NF-κB: nuclear factor-κB; MAP: mitogen-activated protein.
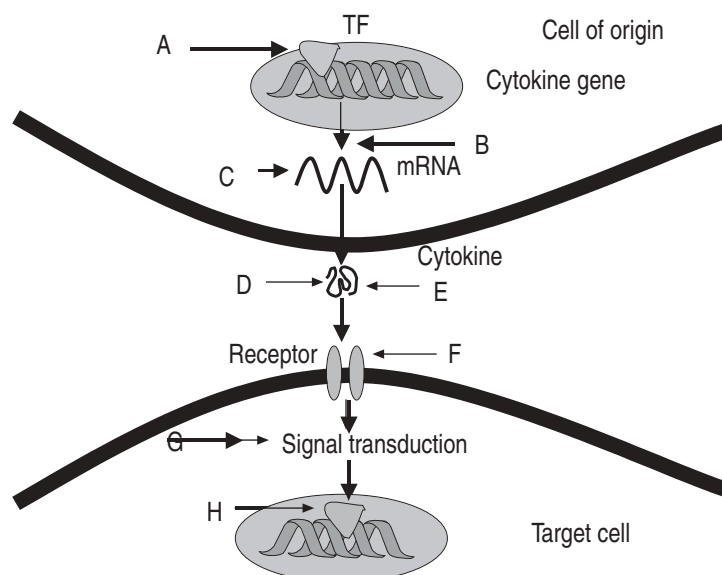
Fig. 1.–Strategies for inhibiting cytokines include inhibition of cytokine synthesis, inhibition of secreted cytokines using blocking antibodies or soluble receptors, and blocking of cytokine receptors and their signal transduction pathways. Horizontal arrows indicate inhibition strategies: A and H: transcription factor (TF) inhibitor; B: synthesis inhibitor; C: antiserve oligonucleotide; D: monoconal antibody; E: soluble receptor; F: receptor antagonist; G: kinase inhibitor. mRNA: messenger ribonucleic acid.

cytokines [6] (fig. 1). Conversely, there are cytokines that themselves suppress the allergic inflammatory process and these may have therapeutic potential in asthma and COPD [8, 9].

Inhibition of Th2 cytokines

Th2-derived cytokines play a key role in orchestrating the eosinophilic inflammatory response in asthma, suggesting that blocking the release or effects of these cytokines may have therapeutic potential. This has been strongly supported by studies in experimental animals, including mice with deletion of Th2-specific cytokine genes. Th2 are unlikely to play any role in COPD and there is no evidence that Th2 cytokine levels are increased in the airways [10, 11].

Anti-interleukin-5

Interleukin (IL)-5 plays an essential role in orchestrating the eosinophilic inflammation of asthma [12, 13]. In IL-5 gene knockout mice, the eosinophilic response to allergen and the subsequent airway hyperresponsiveness (AHR) are markedly suppressed, validating the strategy of inhibiting IL-5 (fig. 2). This has been achieved using blocking antibodies directed against IL-5. These antibodies inhibit eosinophilic inflammation and AHR in animal models of asthma, including primates [14, 15]. This blocking effect may last for up to 3 months after a single intravenous injection of antibody, making treatment of chronic asthma with such a therapy a feasible proposition. Humanized monoclonal antibodies directed against IL-5 have been developed and a single intravenous infusion of one of these antibodies (mepolizumab) markedly reduces blood eosinophil levels for several

weeks and prevents eosinophil recruitment to the airways after allergen challenge in patients with mild asthma [16] (fig. 3). However, this treatment has no significant effect on the early or late response to allergen challenge or on baseline AHR, suggesting that eosinophils may not be of critical importance in these responses in humans. A clinical study in patients with moderate-to-severe asthma, who had not been controlled on inhaled corticosteroid therapy, confirmed a profound reduction in circulating eosinophil numbers, but no significant improvement in either symptoms or lung function [17]. In both of these studies, it would be expected that high doses
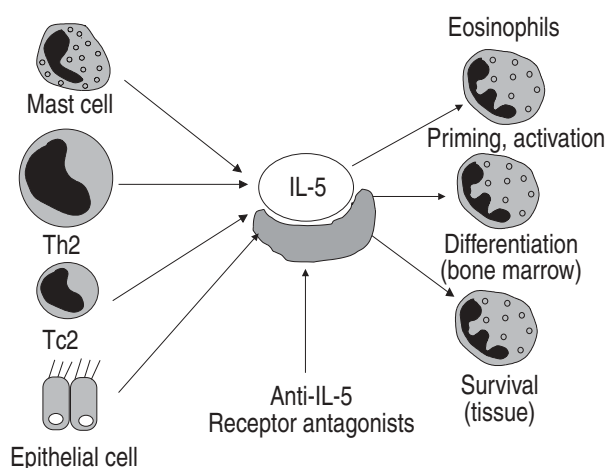

Fig. 2.–Inhibition of interleukin-5 (IL-5). IL-5 is released predominantly from type 2 T-helper lymphocytes (Th2) and the other cells shown and its only effects are on eosinophils, resulting in differentiation in the bone marrow and priming, activation and increased survival in the airways. IL-5 may be blocked using blocking antibodies (such as mepolizumab) or theoretically by receptor antagonists. Tc2: type 2 cytotoxic lymphocyte.
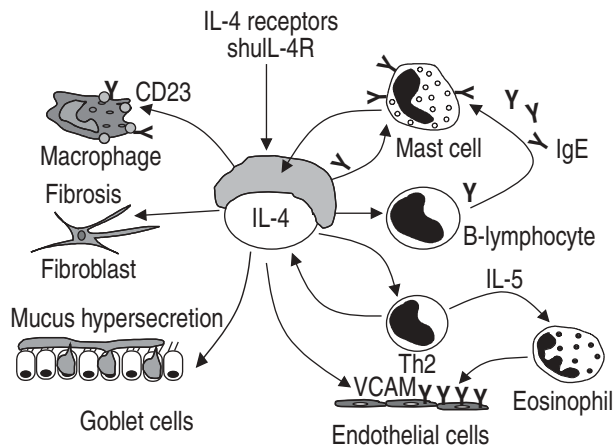
Fig. 3. – Effects of blocking interleukin(IL)-4 in asthma. IL-4 has multiple effects relevant to allergic inflammation in asthma, including differentiation of type 2 T-helper lymphocytes (Th2), production of immunoglobulin E (IgE) from B-lymphocytes, increased expression of the low-affinity receptor for IgE (FCɛRII) on several inflammatory cells, increased mucus secretion and fibrosis. IL-4 may be blocked by a high-affinity soluble receptor (shuIL-4R). VCAM: vascular cell adhesion molecule.

of corticosteroids would improve these functional parameters. These surprising results question the critical role of eosinophils in asthma and indicate that other strategies aimed at inhibiting eosinophilic inflammation might not be effective.

Somewhat similar findings have previously been reported in some studies in mice in which anti-IL-5 reduced eosinophilic responses to allergen, but not AHR, whereas AHR was reduced by anti-CD4, which depletes T-helper cells [18].

The use of nonpeptidic IL-5 receptor (IL-5R) antagonists would be an alternative strategy and such compounds are being sought using molecular modelling of the IL-5R α-chain and through large-scale throughput screening. However, the lack of clinical benefit using anti-IL-5 has made this a less attractive approach. It is possible that eosinophils are associated with more chronic aspects of asthma, such as airway remodelling, and in mice a blocking anti-IL-5 prevents the increased collagen deposition in airways associated with repeated allergen exposure [19]. Eosinophils may be an important source of transforming growth factor-β in asthmatic airways, resulting in structural changes [20].

Anti-interleukin-4

IL-4 is critical to the synthesis of immunoglobulin (Ig) E by B-lymphocytes and is also involved in eosinophil recruitment to the airways [21]. A unique function of IL-4 is to promote differentiation of Th2, and it therefore acts at a proximal and critical site in the allergic response, making IL-4 an attractive target for inhibition (fig. 3).

IL-4-blocking antibodies inhibit allergen-induced AHR, goblet cell metaplasia and pulmonary eosinophilia in a murine model [22]. Inhibition of IL-4 may

therefore be effective in inhibiting allergic diseases, and soluble IL-4Rs are in clinical development as a strategy for inhibiting IL-4. A single nebulized dose of these receptors prevents the fall in lung function induced by withdrawal of inhaled corticosteroids in patients with moderately severe asthma [23]. Subsequent studies have demonstrated that weekly nebulization of the soluble IL-4Rs improves asthma control over a 12-week period [24]. Another approach is blockade of IL-4Rs with a mutated form of IL-4 (BAY 36-1677), which binds to and blocks IL-4Rα and IL-13Rα1, thus blocking both IL-4 and IL-13 [25].

IL-4 and the closely related cytokine IL-13 signal through a shared surface receptor, IL-4Rα, which activates a specific transcription factor, signal transducer and activator of transcription (STAT)-6 [26]. Deletion of the STAT-6 gene has a similar effect to IL-4 gene knockout [27]. This has led to a search for inhibitors of STAT-6, and, although peptide inhibitors that interfere with the interaction between STAT-6 and Janus Kinases linked to IL-4Rα have been discovered, it will be difficult to deliver these intracellularly. An endogenous inhibitor of STATs, suppressor of cytokine signalling-1, is a potent inhibitor of IL-4 signalling pathways and offers a new therapeutic target [26, 28].

Anti-interleukin-13

There is increasing evidence that IL-13 in mice mimics many of the features of asthma, including AHR and mucus hypersecretion, independently of eosinophilic inflammation [29] and potently induces the secretion of eotaxin from airway epithelial cells [30]. IL-13 signals through the IL-4R α-chain, but may also activate different intracellular pathways *via* activation of IL-13Rα1 [31], and thus may be an important target for the development of new therapies. A second specific IL-13R, IL-13Rα2, exists in soluble form and has a high affinity for IL-13, thus acting as a decoy receptor for IL-13. Soluble IL-13Rα2 is effective in blocking the actions of IL-13, including IgE generation, pulmonary eosinophilia and AHR in mice [29]. In the murine model, IL-13Rα2 is more effective than IL-4-blocking antibodies, highlighting the potential importance of IL-13 as a mediator of allergic inflammation. Humanized IL-13Rα2 is now being developed as a therapeutic approach for asthma.

Anti-interleukin-9

IL-9 is a Th2-derived cytokines that may enhance Th2-driven inflammation and amplify mast cell mediator release and IgE production (fig. 4) [32]. IL-9 may also enhance mucus hypersecretion [33]. IL-9 and its receptors show increased expression in asthmatic airways [34, 35]. Strategies for blocking IL-9, including blocking antibodies, are currently in development [36].
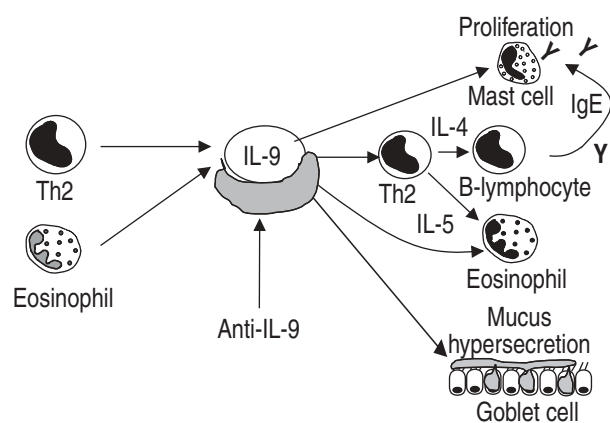
Fig. 4. – Effects of blocking interleukin(IL)-9 in asthma. IL-9 has several effects, including amplification of type 2 T-helper lymphocyte (Th2)-mediated inflammatory effects in the airways. It may be inhibited by a blocking antibody. IgE: immunoglobulin E.

Inhibition of proinflammatory cytokines

Proinflammatory cytokines, particularly IL-1β and tumour necrosis factor-α (TNF-α), may amplify the inflammatory response in asthma and COPD and be linked to disease severity. This suggests that blocking IL-1β or TNF-α may have beneficial effects, particularly in severe airway disease.

Anti-interleukin-1

IL-1 expression is increased in asthmatic airways [37] and activates many inflammatory genes that are expressed in asthma. There are no low-molecular-weight inhibitors of IL-1, but a naturally occurring cytokine, IL-1R antagonist (IL-1RA), binds to IL-1R and blocks the effects of IL-1 [38]. In experimental animals, IL-1RA reduces AHR induced by allergen. Human recombinant IL-1RA does not appear to be effective in the treatment of asthma, however [39]. There are no published studies on the role of IL-1 in COPD.

Anti-tumour necrosis factor-α

TNF-α is expressed in asthmatic airways and may play a key role in amplifying asthmatic inflammation, through activation of nuclear factor-κB (NF-κB), activator protein-1 and other transcription factors [40]. TNF-α levels are markedly increased in induced sputum from patients with asthma [41]. Furthermore, there is evidence that COPD patients with weight loss may show increased releasability of TNF-α from circulating cells and that TNF-α may cause apoptosis of skeletal muscle, resulting in the characteristic cachexia seen in some patients with severe COPD [42]. In rheumatoid arthritis and inflammatory bowel disease, a blocking humanized monoclonal antibody directed against TNF-α (infliximab) and soluble TNF-α receptors (etanercept) have produced remarkable clinical responses, even in patients who are relatively unresponsive to steroids [43, 44]. Such antibodies or soluble TNF-α receptors are a logical approach to asthma therapy, particularly in patients with severe disease, and clinical trials are currently under way. They may also be indicated in the treatment of severe COPD, particularly in patients with malaise and cachexia.

Because of the problems associated with antibody-based therapies, low-molecular-weight inhibitors of TNF-α are being sought. TNF-α-converting enzyme is a matrix metalloproteinase-related enzyme critical for the release of TNF-α from the cell surface. Low-molecular-weight TNF-α-converting enzyme inhibitors are in development as oral TNF-α inhibitors [45].

Anti-inflammatory cytokines

Some cytokines have anti-inflammatory effects in inflammation and therefore have therapeutic potential [8, 9]. Although it may not be feasible or cost-effective to administer these proteins as long-term therapy, it may be possible to develop drugs in the future that increase the release of these endogenous cytokines or activate their receptors and specific signal transduction pathways.

Interleukin-10

IL-10 is a potent anti-inflammatory cytokine that inhibits the synthesis of many inflammatory proteins, including cytokines (TNF-α, granulocyte-macrophage colony-stimulating factor, IL-5 and chemokines) and inflammatory enzymes (inducible nitric oxide synthase) that are overexpressed in asthma (table 2) [46]. Indeed there may be a defect in IL-10 transcription and secretion from macrophages in asthma [47, 48]. In sensitized animals, IL-10 is effective in suppressing the inflammatory response to allergen [49], suggesting that IL-10 might be defective in atopic

Table 2. – The anti-inflammatory actions of interleukin (IL)-10

↓ Proinflammatory cytokines (IL-1β, TNF-α, IL-6, GM-CSF)
↓ Chemokines (IL-8, MIP-1α, RANTES, eotaxin)
↓ Inflammatory enzymes (iNOS, COX-2, MMP-9)
↓ Allergen responses (MHC class II, CD23, B7-1, B7-2)
↓ Th2 cytokines (IL-4, IL-13, IL-5)
↑ Anti-inflammatory effects (IL-1RA, TIMPs)

IL-10 has several anti-inflammatory effects and may therefore be of therapeutic value, via administration of either IL-10 itself (daily subcutaneous injections) and IL-10 analogues or, in the future, drugs that activate the same signal transduction pathways; down arrow: decrease; up arrow: increase; TNF-α: tumour necrosis factor-α; GM-CSF: granulocyte-macrophage colony-stimulating factor; MIP-1α: macrophage inflammatory protein 1α; RANTES: regulated on activation, normal T-cell expressed and secreted; iNOS: inducible nitric oxide synthase; COX-2: cyclo-oxygenase 2; MMP: matrix metalloproteinase; MHC: major histocompatibility complex; Th2: type 2 T-helper lymphocyte; TIMP: tissue inhibitor of MMPs.

diseases. Specific allergen immunotherapy results in increased production of IL-10 by T-helper lymphocytes and this may contribute to the beneficial effects of immunotherapy [50].

IL-10 may also be of therapeutic value in COPD as it inhibits not only TNF-α and chemokines but also certain matrix metalloproteinases (MMPs), such as MMP-9, that may be involved in the destruction of elastin in the lung parenchyma [51]. In addition, IL-10 increases the release of the tissue inhibitors of MMPs, the endogenous inhibitors of MMPs.

Recombinant human IL-10 has proved to be effective in controlling inflammatory bowel disease and psoriasis, in which similar cytokines are expressed, and may be given as a weekly injection [52]. Although IL-10 is reasonably well tolerated, there are haematological side-effects. In the future, drugs which active the unique signal transduction pathways activated by the IL-10R or drugs that increase endogenous production of IL-10 may be developed. In mice, drugs that elevate cyclic adenosine monophosphate increase IL-10 production, but this does not appear to be the case in human cells [53].

Interferons

Interferon gamma (IFN-γ) inhibits Th2 and should therefore reduce atopic inflammation. In sensitized animals, nebulized IFN-γ inhibits eosinophilic inflammation induced by allergen exposure [54]. Administration of IFN-γ by nebulization to asthmatic patients did not significantly reduce eosinophilic inflammation, however, possibly due to the difficulty in obtaining a high enough concentration locally in the airways [55]. Interestingly, allergen immunotherapy increases IFN-γ production by circulating T-lymphocytes in patients with clinical benefit [56] and increased numbers of IFN-γ-expressing cells in nasal biopsy samples from patients with allergic rhinitis [57]. A preliminary report suggests that IFN-α may be useful in the treatment of patients with severe asthma who have reduced responsiveness to corticosteroids [58].

Interleukin-12

IL-12 is the endogenous regulator of Th1 development and determines the balance between Th1 and Th2 [59]. IL-12 administration to rats inhibits allergen-induced inflammation [60] and sensitization to allergens. IL-12 causes IFN-γ release, but has additional effects on T-lymphocyte differentiation. IL-12 levels released from whole blood cells are lower in asthmatic patients, indicating a possible reduction in IL-12 secretion [61].

Recombinant human IL-12 has been administered to humans and has several toxic effects that are diminished by slow escalation of the dose [62]. In patients with mild asthma weekly infusions of human recombinant IL-12 in escalating doses over 4 weeks caused a progressive fall in circulating eosinophil number, and a reduction in the normal rise in circulating eosinophil number after allergen challenge [63]. There was a concomitant reduction in eosinophil number in induced sputum. However, there was no reduction in either the early or late response to inhaled allergen challenge and no reduction in AHR. Furthermore, most of the patients suffered from malaise and one of the 12 subjects had an episode of cardiac arrhythmia. This suggests that IL-12 in not a suitable treatment for asthma. In mice, administration of an IL-12/allergen fusion protein results in the development of a specific Th1 response to the allergen, with increased production of an allergen-specific IgG2, rather than the normal Th2 response with IgE formation [64]. This indicates the possibility of using localized IL-12 together with specific allergens to provide a more specific immunotherapy, which might even be curative if applied early on in the course of the atopic disease.

Interleukin-18

IL-18 was originally described as IFN-γ-releasing factor, but has a different mechanism of action to IL-12 [65]. IL-12 and IL-18 appear to have a synergistic effect in inducing IFN-γ release and inhibiting IL-4-dependent IgE production and AHR [66].

Chemokine inhibitors

Many chemokines are involved in the recruitment of inflammatory cells in asthma and COPD. >50 different chemokines that activate up to 20 different surface receptors are now recognized [67, 68]. Chemokine receptors belong to the seven-transmembrane-spanning domain receptor superfamily of G-protein-coupled receptors and this makes it possible to find low-molecular-weight inhibitors, which has not been possible for classical cytokine receptors [69]. Some chemokines appear to be selective for single chemokines, whereas others are promiscuous and mediate the effects of several related chemokines (table 3). Chemokines appear to act in sequence in determining the final inflammatory response and so inhibitors may be more or less effective depending on the kinetics of the response [70].

CCR3 inhibitors

Several chemokines, including eotaxin, eotaxin-2, eotaxin-3, regulated on activation, normal T-cell expressed and secreted (RANTES) and macrophage chemoattractant protein (MCP)-4, activate a common receptor on eosinophils termed CCR3 [71]. A neutralizing antibody directed against eotaxin reduced eosinophil recruitment to the lung after allergen challenge and the associated AHR in mice [72]. There is increased expression of eotaxin, eotaxin-2, MCP-3, MCP-4 and CCR3 in the airways of asthmatic patients and this correlates with increased AHR [73, 74]. Several long-molecular-weight inhibitors of CCR3, including UCB 35625, SB297006 and SB328437, are effective in inhibiting eosinophil

Table 3. – Chemokine receptor antagonists in asthma

Chemokine receptor	Cell types	Agonists	Antagonists
CCR3	Eosinophil, Th2, mast cell	Eotaxin, eotaxin-2, RANTES, MCP-4	Met-RANTES, UCB 35625, SB 328437
CCR2	Monocyte, mast cell, T-lymphocyte	MCP-1, MCP2-5	
CCR4	Th2	MDC, TARC	

Several chemokines are likely to be involved in the patho-physiology of asthma. There are three major chemokine receptor targets in asthma, CCR3, which is most advanced in terms of low-molecular-weight inhibitor development, and also CCR2 and CCR4, for which low-molecular-weight inhibitors are currently developed; Th2: type 2 T-helper lymphocyte; RANTES: regulated on activation, normal T-cell expressed and secreted; MCP-4: macrophage chemoattractant protein-4; MDC: monocyte-derived chemokine; TARC: thymus and activation-dependent chemokine; Met-RANTES: N-terminally modified RANTES.

recruitment in allergen models of asthma [75, 76] and drugs in this class are currently undergoing clinical trials in asthma. Although it was thought that CCR3 were restricted to eosinophils, there is some evidence for their expression on Th2 and mast cells; thus, these inhibitors may have a more widespread effect than on eosinophils alone, making them potentially more valuable in asthma treatment.

RANTES, which shows increased expression in asthmatic airways [77] also activates CCR3, as well as having effects on CCR1 and CCR5, which may play a role in T-lymphocyte recruitment. Modification of the N-terminus of RANTES, to give met-RANTES, has a blocking effect on RANTES by inhibiting these receptors [78].

CCR2 inhibitors

MCP-1 activates CCR2 on monocytes and T-lymphocytes. Blocking MCP-1 with neutralizing antibodies reduces recruitment of both T-lymphocytes and eosinophils in a murine model of ovalbumin-induced airway inflammation, with a marked reduction in AHR [72]. MCP-1 also recruits and activates mast cells, an effect that is mediated *via* CCR2 [79]. MCP-1 instilled into the airways induces marked and prolonged AHR in mice, associated with mast cell degranulation. A neutralizing antibody to MCP-1 blocks the development of AHR in response to allergen [79]. MCP-1 levels are increased in the bronchoalveolar lavage fluid of patients with asthma [80]. This has led to a search for low-molecular-weight inhibitors of CCR2.

CCR2 may also play an important role in COPD, as MCP-1 levels are increased in the sputum and lungs of patients with COPD [81, 82]. MCP-1 is a potent chemoattractant of monocytes and may therefore be involved in the recruitment of macrophages in COPD.
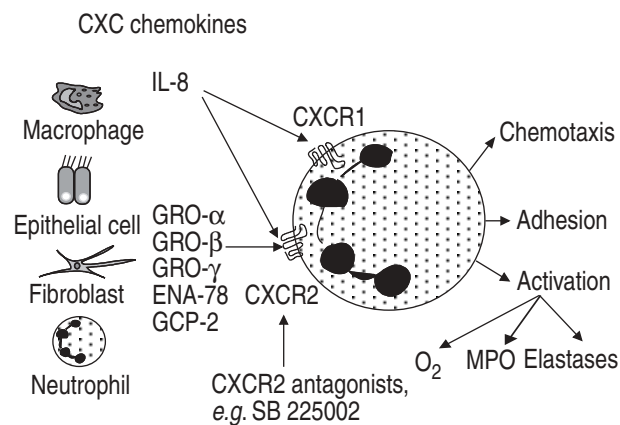

Fig. 5. – CXC chemokine receptors (CXCRs) are important in neutrophilic inflammation. Interleukin-8 (IL-8) signals through a specific low-affinity receptor (CXCR1) and also through a high-affinity receptor (CXCR2) shared with other CXC chemokines, such as growth-related oncogene (GRO)-α. Low-molecular-weight inhibitors of CXCR2 are curretly developed. The cells depicted at the left are the sources of CXC chemokine. ENA-78: epithelial cell-derived neutrophil-activating peptide-78; GCP-2: granulocyte chemotactic protein-2; MPO: myeloperoxidase.

Indeed, the chemoattractant effect of induced sputum from patients with COPD is abrogated by an antibody to CCR2 [82]. Since macrophages appear to play a critical role in COPD as a source of elastases and neutrophil chemoattractants, blocking CCR2 may be important as a therapeutic strategy in COPD.

CCR4 inhibitors

CCR4 are selectively expressed on Th2 and are activated by the chemokines monocyte-derived chemokine and thymus and activation-dependent chemokine [83]. Inhibitors of CCR4 may therefore inhibit the recruitment of Th2 and thus cause persistent eosinophilic inflammation of the airways.

CXC receptor inhibitors

CXC receptors mediate the effects of CXC chemokines, which act predominantly on neutrophils. IL-8 levels are markedly increased in the induced sputum of patients with COPD and correlate with the increased proportion of neutrophils [41, 84]. Since IL-8 is a potent neutrophil chemoattractant, it is an attractive target for COPD therapy. Anti-IL-8 has an inhibitory effect on the chemotactic response to COPD sputum [85]. IL-8 acts *via* two receptors, CXCR1, which is a low-affinity receptor that is specific for IL-8, and CXCR2, which has high affinity and is shared by several other CXC chemokines, including growth-related oncogene (GRO)-α, GRO-β, GRO-γ, granulocyte chemotactic protein-2 and epithelial cell-derived neutrophil-activating peptide-78 (fig. 5). CXCR1 responds to high concentrations of IL-8 and is responsible for activation of neutrophils and release of superoxide anions and neutrophil elastase, whereas CXCR2 responds to low concentrations of CXC

chemokines and is involved in chemotactic responses. Potent low-molecular-weight inhibitors of CXCR2 that block the chemotactic response of neutrophils to IL-8 and GRO-α, such as SB 225002, have now been developed [86]. This antagonist has a significant inhibitory effect on the chemotactic response to COPD sputum. Concentrations of GRO-α are also elevated in the induced sputum of patients with COPD and this mediator has a chemotactic effect on neutrophils and monocytes [82]. CXCR2 antagonists may therefore also reduce monocyte chemotaxis and the accumulation of macrophages in COPD patients.

Neutrophils are not a prominent feature of inflammation in patients with chronic asthma and inflammation is dominated by eosinophils. However, there is evidence for increased neutrophils number in the biopsy samples and induced sputum of patients with severe asthma who are treated with high doses of inhaled or oral corticosteroids, and levels of IL-8 are increased [87, 88]. It is not certain whether this neutrophilic inflammation contributes to the asthma pathophysiology, but it is possible that CXCR inhibitors may have a therapeutic role in severe asthma.

Other approaches to cytokine inhibition

Although several attempts have been made to block specific cytokines, this may not be adequate to combat the chronic inflammation in asthma and COPD as so many cytokines are involved and there is considerable redundancy of effects. This has led to the suggestion that development of drugs that have a more general effect on cytokine synthesis may be more successful. However, these drugs also affect other inflammatory processes, and so their beneficial effects cannot necessarily be ascribed to inhibition of cytokine synthesis alone.

Corticosteroids

Corticosteroids represent the most effective treatment for asthma by far and part of their efficacy is due to inhibition of inflammatory cytokine expression. This is mediated *via* an effect on glucocorticoid receptors, reversing the acetylation of core histones that is linked to increased expression of inflammatory genes [89]. However, corticosteroids are not effective in suppressing the inflammation in COPD [90] and this, at least in part, may be explained by an inhibitory effect of cigarette smoking on histone deacetylation [91].

Immunomodulatory drugs

Ciclosporin A, tacrolimus and rapamycin inhibit the transcription factor nuclear factor of activated T-cells that regulates the secretion of IL-2, IL-4, IL-5 and granulocyte-macrophage colony-stimulating factor by T-lymphocytes [92]. Although it has some reported beneficial steroid-sparing effects in asthma

[93], the toxicity of ciclosporin A limits its usefulness, at least when given orally. More selective Th2-selective drugs may be safer for the treatment of asthma in the future. An inhibitor of Th2-derived cytokines, suplatast tosilate [94], is reported to provide clinical benefit in asthma [95]. Cytotoxic (CD8+) T-lymphocytes are prominent in COPD and therefore immunomodulatory drugs may also have a role in this disease.

Phosphodiesterase 4 inhibitors

Phosphodiesterase 4 (PDE_4) inhibitors inhibit the release of cytokines and chemokines from inflammatory cells *via* an increase in intracellular cyclic adenosine monophosphate [96]. Their clinical use is limited in asthma by side-effects such as nausea. In contrast to corticosteroids, PDE_4 inhibitors have a potent inhibitory effect on neutrophils [97], indicating that they may be useful anti-inflammatory treatments for COPD. There is preliminary evidence that a PDE_4 inhibitor cilomilast improves lung function and symptoms in patients with COPD, although whether this is due to inhibition of cytokines is not yet certain [98].

Nuclear factor-κB inhibitors

NF-κB regulates the expression of many cytokines and chemokines involved in asthma and COPD [99]. There are several possible approaches to inhibition of NF-κB, including gene transfer of the inhibitor of NF-κB (IκB), inhibitors of IκB kinases, NF-κB-inducing kinase and IκB ubiquitin ligase, which regulate the activity of NF-κB, and the development of drugs that inhibit the degradation of IκB [100]. One concern about this approach is that effective inhibitors of NF-κB may result in immune suppression and impair host defences, since knockout mice, which lack NF-κB proteins, succumb to septicaemia. However, there are alternative pathways of NF-κB activation that might be more important in inflammatory disease [101].

p38 mitogen-activated protein kinase inhibitors

Mitogen-activated protein (MAP) kinases play a key role in chronic inflammation and several complex enzyme cascades have now been defined. One of these, the p38 MAP kinase pathway, is involved in the expression of inflammatory cytokines and chemokines [102, 103]. Nonpeptide inhibitors of p38 MAP kinase such as SB 203580, SB 239063 and RWJ 67657, also known as cytokine synthesis anti-inflammatory drugs, have now been developed and these drugs have a broad range of anti-inflammatory effects [104]. However, there may be issues of safety as p38 MAP kinases are involved in host defence. It is possible that the inhalational route of delivery will reduce the risk of side-effects.

Conclusions

Several specific cytokine and chemokine inhibitors are currently being developed for the treatment of asthma and COPD.

Inhibition of IL-4 with soluble IL-4 receptors has shown promising early results in asthma. Anti-IL-5 is very effective at reducing peripheral blood and airway eosinophil numbers, but does not appear to be effective against symptomatic asthma. Inhibitory cytokines, such as IL-10, interferons and IL-12 are less promising, as systemic delivery causes side-effects, and it may be necessary to develop inhalation delivery systems. Inhibition of TNF-α may be useful in the treatment of severe asthma and COPD.

Many chemokines are involved in the inflammatory response of asthma and COPD and low-molecular-weighted inhibitors of chemokine receptors are currently in development. CCR3 and CXCR2 antagonists are also currently being developed for the treatment of asthma and COPD respectively.

Because so many cytokines are involved in these complex diseases, drugs that inhibit the synthesis of multiple cytokines may be more successful. Several such classes of drug are now in clinical development. The risk of side-effects in these nonspecific inhibitors may be reduced by use of inhalational route of delivery, however.

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