

EDITORIAL

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Airway smooth muscle: telling the eosinophil how to behave

This editorial discusses the findings of the paper in this issue by Fanat et al. [6] pp. 1009–17.

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It is over 20 years since the discovery of interleukin-5 (IL-5) and the demonstration of its role as an eosinophil differentiation and chemotactic factor [1, 2]. A few years before the discovery of IL-5, the evidence supporting the importance of the eosinophil in the pathogenesis of bronchial asthma was beginning to mount [3]. Initially reported as an eosinophil colony stimulating factor (Eo-CSF), the ability of IL-5 to impact on the behaviour and survival of mature eosinophils and indeed to be produced by eosinophils themselves has led to an avalanche of research into its potential as a therapeutic target in asthma and allergic inflammation.

The view of airway smooth muscle (ASM) as merely a contractile tissue, hyperresponsive in asthmatic patients, is a thing of the past. There is an increasing body of evidence supporting the importance of an ASM-derived chemokine/cytokine component in allergic asthma [4]. Numerous molecules released by ASM act as chemoattractants for inflammatory cells (e.g. IL-8 as a neutrophil attractant) and amplifiers of signals released by such cells. Rather than a strict IL-8 = neutrophil attractant; IL-5/eotaxin = eosinophil attractants, there appears to be cross-over in the ability of these molecules to attract specific cell types. Given the increased production of progenitor cells [eosinophil/basophil colony forming units (Eo/B CFUs)] by the bone marrow of atopic asthmatics receiving an allergen challenge [5], it stands to reason that a mechanism for recruitment to and maturation of these cells in the lungs exists. In this issue of the journal Fanat and

colleagues' *in vitro* experiments suggest that stimulated ASM may produce the chemokines/cytokines necessary for the recruitment and differentiation of these cells [6].

Fanat and colleagues used a cocktail of pro-inflammatory cytokines to stimulate ASM cultured from *non-asthmatic* lung tissue. In previous studies, similar stimulation has been shown to cause the production of eotaxin (also an attractant of mast cells) [7], RANTES (regulated upon activation, normal T cell expressed and secreted) [8] (a potent attractant of monocytes, memory T cells, and eosinophils), IL-8 [9], monocyte chemoattractant proteins, stem cell factor [10], granulocyte macrophage colony stimulating factor (GM-CSF) [11], and IL-6 [12]. So while the growth and differentiation of progenitor cells into mature eosinophils in the presence of supernatant from ASM after this stimulation is novel, it is perhaps not surprising. No difference was shown between the numbers of eosinophil colonies grown from the atopic group compared with normal but perhaps this was due to mild disease. Replication of the experiments in a group of atopic patients 24-h post-allergen challenge may produce different results.

There is conflicting evidence concerning the cytokines responsible for the maturation of progenitor cells to mature inflammatory cells in the lung with GM-CSF and IL-5 both being implicated. Fanat and colleagues show the application of IL-5 antibodies ablated the eosinophilopoietic potential of the stimulated ASM and while somewhat tangential to the current article, the recent publication of a positive clinical trial of Mepolizumab (a humanized monoclonal antibody against IL-5) in refractory eosinophilic asthma [13] highlights the clinical potential of intervention in this cascade. This clinical trial showed a significant reduction in exacerbations in the treatment arm in a group of exacerbation-prone patients, highlighting the importance of IL-5-mediated/propagated eosinophilic inflammation in the acute worsening of disease in this group. It is 9 years since the failure of an

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IL-5 monoclonal antibody (SB 240563) to lessen airway hyperresponsiveness and late asthmatic response was reported in a clinical trial despite a potent reduction in blood and sputum eosinophils [14]. The population used in the recent trial had sputum eosinophilia despite corticosteroid treatment, highlighting the importance of detailed phenotyping of 'difficult asthma' patients.

Fanat and colleagues also investigated the relative importance of three classical signalling pathways involved in ASM stimulated eosinophilopoiesis. In their experiments non-cytotoxic levels of typical and atypical isoforms of protein kinase C (PKC) antagonists (Calphostin and Rotlerin), respectively, ablated and attenuated differentiation stimulated by both IL-5 and the ASM supernatant. In addition, modulation of the IL-5-mediated p38 MAP kinase pathway was shown to have a similar effect. However, inhibition of the src tyrosine kinase pathway proved to be ineffective in attenuating differentiation and the kinase was not detectable in lysates of CD34⁺ cells co-cultured with IL-5 and GM-CSF. Further work is required to elucidate the role of the PKC isoforms but their results provide an insight into the IL-5-like signalling pathway used by ASM to stimulate eosinophil differentiation; that is to say dependent on p38 MAP kinase but not src tyrosine kinase. These results suggest that approaches targeting p38 MAP kinase may have a therapeutic role in disrupting smooth muscle–eosinophil interactions in asthma.

The relative role played by ASM compared to other cell types in the theatre of pulmonary cytokine production *in vivo* is difficult to quantify. However, the large ASM mass and high concentrations of inflammatory molecules produced suggests that it is likely to be a major if not the major player. Furthermore, ASM from asthmatic subjects responds differently, both qualitatively and quantitatively compared with normal subjects. *In vitro* experiments using *ex vivo* tissue from well-characterized subjects may lead us a step closer to a more comprehensive understanding of the underlying pathophysiological mechanisms in this disease spectrum.

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