

EDITORIAL

Haemopoietic mechanisms in allergic rhinitis

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In the adult, haemopoietic stem cell differentiation and maturation have traditionally been thought to be restricted to the bone marrow microenvironment. However, a novel view has emerged in recent years according to which at least some haemopoietic (and non-haemopoietic) stem cells present in adult tissue may be recruited from the bone marrow, through the peripheral circulation, into mucosal tissues, becoming part of a regenerative and/or inflammatory process at 'distal' tissue sites. This process has been referred to as the *plasticity* of stem cells [1]. More specifically, haemopoietic progenitor cells have the potential not only to give rise to mature cells within the bone marrow, which can then egress into the circulation, but may themselves also egress from the bone marrow, and home to various organs and tissues under the orchestrated control of specific chemokines and cytokines. Once within the tissue, the fate of these primitive haemopoietic progenitor cells is determined by locally elaborated growth factors that permit a process termed '*in situ* haemopoiesis' [2–5]. We provide herein an overview of the role of systemic and *in situ* haemopoiesis, as components of the already complex scenario of allergic inflammation within the airways in allergic rhinitis (AR) and related disorders.

A considerable body of evidence now exists showing that activation of selective haemopoietic processes is not only associated with the onset and maintenance of allergic inflammation in atopic adults but also with the development of allergic diathesis in infants. Using antibodies in a haemopoietic progenitor membrane protein marker, CD34, and in haemopoietic cytokine receptors, we have demonstrated alterations relevant to eosinophil/basophil lineage commitment of progenitors in neonates at risk for atopy [6, 7]. CD34 is a stage-specific antigen, which, used in combination with small-size and low-granularity determinants, can accurately enumerate progenitor cells in cord blood, peripheral blood, bone marrow and, more recently, lung tissue and sputum by flow cytometric methodologies [8–11]. The CD34 antigen is also expressed on tissue structural cells, including fibroblasts and vascular endothelial cells, and thus caution should be exercised when enumerating progenitor cells by single-stain immunocytochemistry [12, 13]. However, the use of double-staining techniques where CD34⁺ cells are co-stained with CD133, expressed by primitive haemopoietic stem and progenitor cells and retinoblastoma [14], may provide more accurate estimates of progenitor cells within human tissue. The exact role of CD34 has not yet been elucidated, although it has been suggested that it maintains stem cells in the G0 phase [15]. Properties as an adhesion ligand for L-selectin are now known to be restricted to CD34 expressed on high-vein endothelial cells in lymph nodes only and not to haemopoietic CD34 [16]. Although the expression of CD34 has recently been described on immature but not

mature eosinophils [17], its role in the maturation and migrational responses of these cells remains to be clarified. This is particularly important in light of studies with CD34 knockout mouse models, in which in only one of two reports was there a down-modulation of leucocyte trafficking [18] and a reduction of numbers of myeloid progenitor cells [19].

Studies in adult subjects have shown that circulating, common progenitors of eosinophils and basophils, measured as *in vitro* colony-forming units (Eo/B-CFU), are detected in greater numbers in the peripheral blood of asymptomatic atopics, compared with non-atopic controls; these Eo/B-CFU are responsive to activated T cell supernatants and, specifically, to IL-5 [8, 20]. Additional studies extended these findings by showing relevant fluctuations in blood and bone marrow Eo/B-CFU in response to allergen-induced allergic inflammatory responses [21–23]. Of note, the finding that following natural allergen exposure, the numbers of circulating Eo/B-CFU were greatly reduced in symptomatic AR at the peak of a pollen season was the first indication that, during the inflammatory process, progenitor cells may home to tissue site(s) of inflammation [24–27]. In addition, the detection of locally elaborated haemopoietic growth factors and Eo/B-CFU within nasal polyps further supported the concept of *in situ* haemopoiesis [5, 28]. This view was strengthened by findings that: (i) CD34-immunopositive/IL5R α mRNA⁺ cells were detected in lung biopsies from atopic asthmatics [29]; (ii) *ex vivo* allergen challenge of nasal explant tissue from AR demonstrated IL-5-driven eosinophil differentiation [30]; and (iii) in mouse models of allergen-induced airway eosinophilia, increased numbers of IL-5-responsive Eo/B-CFU could be grown from lung-extracted progenitors following allergen, compared with saline challenge [11].

Mouse models of AR have highlighted the multifactorial nature of this inflammatory disease, and directly implicate the role of haemopoietic processes in allergic airways disease, specifically in AR. The up-regulation of myeloid progenitors in the bone marrow after airway allergen inhalation [31, 32], and their trafficking to the airways from the marrow in several animal models of either upper or lower airways inflammation have been demonstrated [33–35]. The resultant blood, nasal and/or pulmonary eosinophilia in these models can be blocked by antibodies to IL-5 [36], or by deletion of the gene encoding IL-5 [37, 38]; eotaxin is also critical in this process [9, 39]. Rather than demonstrating a complete ablation of the allergic inflammatory response in the nose, IL-5-deficient mice develop delayed nasal symptoms and basophilic, rather than eosinophilic, inflammation within the nasal mucosa [32, 33]. Haemopoietic cytokine redundancy among IL-3, IL-5 and granulocyte macrophage-colony stimulating factor is thought to ensure adequate production

of inflammatory cell progenitors even in the absence of one of these cytokines, e.g. IL-5 [35]. These data support the concept that systemic, haemopoietic mechanisms are involved in rhinitis as well as asthma.

Phenotypic analyses of progenitor cells using CD34, together with antibodies to cytokine and chemokine receptors, have provided an insight into the contribution of systemic and tissue related haemopoietic processes in the development of allergic inflammatory responses. We have shown that, whereas up-regulation of IL-5R α on CD34⁺ cells within the bone marrow promotes increased eosinophilopoiesis [9], the up-regulation of CCR3 on CD34⁺ cells may favour increased traffic of progenitor cells from the bone marrow to the peripheral circulation in response to eotaxin [40]. Chemokines may therefore orchestrate the egress of progenitor cells from the bone marrow microenvironment and the homing of progenitor cells to tissue sites of inflammation. An important candidate may be stromal cell-derived factor-1 (SDF-1), which is a potent progenitor cell chemoattractant and plays a role in stem cell homing during embryogenesis [41].

In the present issue of the journal, two papers give us further insight into the role of progenitor cells in AR, and specifically to events that may be occurring in the tissue in response to systemic perturbations. In the study by Sergejeva et al. [42] using nasal biopsy tissue from AR, immunopositive CD34 cells have been detected in nasal biopsy tissue; changes in numbers of these cells in relation to natural allergen exposure are discussed. The authors demonstrate increases of immunopositive CD34 cells and immunopositive CD34 cells co-expressing CXCR4, as well as eosinophils expressing CD34, following single-allergen challenge in nasal biopsy. Although these data suggest that SDF-1 may play a role in recruiting progenitor cells into the inflamed tissue, further studies are required to verify this. Interestingly, in the group of subjects who were pre-treated with fluticasone compared with placebo, this study showed a significant reduction in CD34 cells and immunopositive CD34 cells co-expressing CXCR4, as well as eosinophils expressing CD34, following allergen exposure. This is contrary to a previous study with nasal polyp tissue in which topical steroid pre-treatment significantly increased CD34⁺ cell numbers but reduced eosinophilia indicating the down-modulating effect of topical corticosteroids on *in situ* eosinophilopoiesis [28].

In a second study related to this topic in the current issue, Wilson et al. [43] have shown that, in subjects with AR and pre-existent sputum eosinophilia in whom a systemic, haemopoietic process may be expected to be responsive to, and up-regulated by, allergen challenge in the nose [44], a single-allergen nasal challenge provoked increases in nasal symptoms and eosinophilia. In contrast, however, to the mouse models referred to above and to previous data in seasonal AR, no changes were detected in the numbers of Eo/B-CFU 24 h post-allergen. As discussed by the authors, the lack of relevant fluctuations in progenitor cell populations within the blood may reflect lack of adequate powering of the study. In addition, based on recent data we have acquired in our experimental AR and airway eosinophilia models, the time course of blood sampling may have missed the peak of Eo/B-CFU changes in response to allergen: kinetic studies in the mouse model have shown changes in eosinophilopoiesis as

early as 2 h after allergen challenge, indicating that progenitor cell fluctuations may occur more rapidly than originally postulated [11, 45].

The activation of eosinophilopoietic processes within haemopoietic compartments, and, importantly, at mucosal tissue sites during an allergic inflammatory response, provides novel targets for the treatment of allergy as a systemic process and disease. The clinical limitations of monotherapy with topical corticosteroids, oral cysteinyl leukotriene antagonists and cytokine antagonists such as antibodies to IL-5 [46], among others, point out that suppression of haemopoietic contributions to allergic inflammation may constitute a prerequisite for full control of allergic inflammation and disease [4, 27]. The papers in this issue emphasize the importance of haemopoietic processes in the development of AR, and, by extension, in its optimal control.

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