EDITORIAL

Mast cell regulation of airway smooth muscle function in asthma

P. Bradding

Asthma is a common disease affecting $\leqslant 10\%$ of the adult Western population [1, 2]. It is characterised by the presence of variable and potentially reversible airflow obstruction, which occurs as the result of bronchoconstriction, airway mucus plugging and airway oedema. Pathologically there is evidence of airway inflammation and remodelling [3–7]. The mucosal inflammatory infiltrate commonly comprises activated T-cells, eosinophils and mast cells, while the accompanying structural changes include subepithelial collagen deposition, goblet cell- and mucous gland-hyperplasia, airway smooth muscle (ASM) hypertrophy and ASM hyperplasia.

An important physiological feature of asthma is the presence of bronchial hyperresponsiveness (BHR) [8]. This means there is an exaggerated bronchoconstrictor response of the ASM to direct and indirect stimuli such as histamine and exercise, respectively. While BHR is aggravated in the presence of classic allergic eosinophilic airway inflammation, it persists once this inflammation is controlled and is not present in patients with eosinophilic bronchitis [8, 9]. This suggests that there is either a fundamental abnormality of ASM behaviour in asthmatic subjects or that there are interacting factors that have not been previously recognised. In support of the former, several phenotypic differences are evident in ASM cells cultured from the airways of asthmatic subjects. For example, when compared with normal ASM cells, cultured asthmatic ASM cells proliferate faster due to an altered pattern of matrix protein deposition [10, 11], secrete greater amounts of connective tissue-derived growth factor in response to transforming growth factor-β stimulation [12], and secrete markedly increased amounts of the chemokine CXCL10 in response to activation by cytokines [13]. There is decreased expression of prostaglandin (PG)E_2 by the asthmatic ASM [14], and proliferation of asthmatic ASM is not inhibited by corticosteroids due to impaired expression of the transcription factor CCAAT/enhancer binding protein-α [15]. It is remarkable that these differences are evident after several passages in culture; moreover, these differences support the view that there is, in part, a primary ASM abnormality in asthma.

Mast cells play a significant role in the pathophysiology of asthma due to their ability to release a host of pleiotropic autacoid mediators, proteases and cytokines in response to activation by both immunoglobulin (Ig)E-dependent and diverse nonimmunological stimuli [16, 17]. For example, following laboratory allergen challenge, secretion of the autacoid mediators histamine, PGD_2 and leukotriene (LT)C_4 induces bronchoconstriction, mucus secretion and mucosal oedema, thus contributing to acute symptoms. Mast cell-derived cytokines include interleukin (IL)-4, IL-5 and IL-13, which regulate both IgE synthesis and the development of eosinophilic inflammation [18]. In addition, the mast cell neutral proteases, tryptase and chymase, interact with many cells that potentially contribute to airway wall remodelling. Importantly, in chronic asthma, mast cells within the bronchial mucosa are in an ''activated'' secretory state, with evidence of ongoing mediator release and cytokine synthesis [19–22].

Mast cells are found adjacent to blood vessels in the lamina propria in normal human airways, but in asthma they migrate into three key structures: the airway epithelium [23]; the airway mucous glands [24]; and the ASM [25]. This anatomical relocation places the mast cell within several dysfunctional airway elements and suggests that the targeted delivery of their mediators is likely to be central to the disordered airway physiology. Of particular interest in relation to bronchoconstriction and BHR is the presence of mast cells within the ASM bundles. Eosinophilic bronchitis is a common cause of cough and is characterised by the presence of a sputum eosinophilia occurring in the absence of variable airflow obstruction or BHR [9]. A detailed comparison of the immunopathology of asthma and eosinophilic bronchitis has revealed an identical pathology in terms of T-cell infiltration, activation status and phenotype, eosinophil infiltration and activation, mucosal mast cell numbers, T-helper cell (Th)2 cytokine expression, epithelial integrity, sub-basement membrane collagen deposition and mediator concentrations, including histamine and PGD_2 [25–27]. This suggests that many of the immunopathological features previously attributed as causing asthma may not be as important for the development of airflow obstruction, BHR and remodelling as previously suggested. The striking difference between the pathology of asthma and eosinophilic bronchitis resided within the ASM bundles, which contained numerous mast cells in asthma patients but virtually none in normal subjects or patients with eosinophilic bronchitis [25]. The majority of these mast cells contained both tryptase and chymase, and expressed IL-4 and IL-13 but not IL-5. Interestingly, there were almost no T-cells or eosinophils in the smooth muscle of any of the study groups. There was a significant correlation between ASM mast cell number and

CORRESPONDENCE: P. Bradding, Dept of Respiratory Medicine, Glenfield Hospital, Groby Rd, Leicester, LE3 9QP, UK. Fax: 44 1162502787. E-mail: pbradding@hotmail.com

BHR within the asthmatic group supporting the view that this observation is of functional relevance. Such initial findings have been confirmed by several independent groups [28–31] and it has recently been shown that the mast cells within the ASM bundles in asthma demonstrate ultrastructural features of activation [31]. These studies suggest that ASM infiltration by mast cells is one of the critical determinants of the asthmatic phenotype [28–31].

The specific recruitment of mast cells to the ASM in asthma raises several important questions; in particular, whether the cells interact, and if so, what are the functional consequences for airway function? Studies examining whole cell interactions in vitro are sparse. However, human lung mast cells adhere to ASM cells, in part, via an interesting molecule known as tumour suppressor in lung cancer-1, suggesting that specific cellular cross-talk is likely [32]. In addition, potential mechanisms of mast cell recruitment by the ASM have been identified [13] and inhibition of this recruitment may offer a new angle on asthma therapy [33]. In contrast, it has been known for several years that numerous mast cell-derived mediators directly affect ASM function when examined in isolation. For example, the mast cell autacoid mediators histamine, PGD_2 and LTC_4 are all potent agonists for ASM contraction, and exogenously administered tryptase induces bronchoconstriction and BHR in response to histamine in dogs and sheep [34, 35]. In vitro, tryptase potentiates the contractile response of sensitised bronchi to histamine [36] and induces proliferation of human ASM [37, 38]. Instillation of Th2 cell conditioned medium to the airways of naïve mice induces BHR within 6 h, and requires expression of the IL-4 receptor α-subunit and signal transducer and activator of transcription (STAT)6, suggesting a critical role for IL-4 and/or IL-13. Both of these ILs produce similar effects when administered individually [39]. TNF-α induces BHR in normal subjects and exacerbates BHR in patients with asthma [40, 41], while blocking tumour necrosis factor (TNF)-α activity in asthma patients improves BHR [42, 43]. Human lung mast cells are a source of IL-4, IL-13 and TNF-α [23, 44–46], which suggests a further mechanism through which these cells could contribute to the development of BHR.

In the present issue of the European Respiratory Journal, CHHABRA et al. [47] have examined the ability of two mast cell mediators, histamine and tryptase, to modify the synthetic ability of the ASM derived from both asthmatic and nonasthmatic subjects [47]. Specifically, they examined the release of the cytokine granulocyte-macrophage colony-stimulating factor (GM-CSF) and the RANTES (regulated on activation, normal T-cell expressed and secreted) chemokine. The nonasthmatic subjects were patients undergoing lung transplantation for a variety of end-stage lung diseases or resection for carcinoma, while the asthmatic ASM was obtained at bronchoscopy. However, no important differences in secretory responses were found between the two groups, so it is unlikely that any asthma-specific effects were missed. The main observation from the study was that histamine potentiated IL-1β-induced GM-CSF production but inhibited TNF-α-induced RANTES production. In contrast, tryptase only increased GM-CSF secretion after stimulation by both IL-1β and TNF-α, and did not affect RANTES secretion. Pharmacological blockade suggested that the effects of histamine were predominantly mediated via the

H1 and not the H2 receptor. This was unexpected as CHHABRA et al. [47] predicted that stimulation of H2 receptors would increase intracellular cyclic adenosine monophosphate and thus inhibit GM-CSF release, while at the same time potentiating RANTES release. The authors therefore proposed that the H1-mediated effects predominated and occur via activation of H1-coupled phospholipase C. However, it must be borne in mind that G-protein coupled receptors (GPCRs), such as H1 and H2, that were once thought to mediate all their effects via modulation of intracellular cyclic nucleotides are now known to couple to many diverse signalling pathways [48]. For example, these include the membrane-delimited modulation of numerous ion channels involved in the regulation of intracellular calcium signals [49, 50].

The biological significance of the observations of CHHABRA et al. [47] is uncertain. They highlight how the behaviour of cells cannot always be readily predicted and that the potential effects of mast cells on ASM function are undoubtedly complex. It is possible that the effects of histamine and tryptase in combination may differ from those of the individual mediators alone, for example via GPCR cross-talk, and so it would have been interesting for the authors to also examine this. It is also possible that the complex milieu of mediators released from intact mast cells will produce yet different effects, so in the future it will be important to study these cells in co-culture.

In summary, the location of mast cells within the airway smooth muscle bundles is likely to be important for the pathophysiology of asthma and may occur in response to, with subsequent aggravation of, an underlying abnormality in the behaviour of asthmatic airway smooth muscle. Understanding the consequences of this cellular interaction through the type of study by CHHABRA et al. [47] may offer new approaches to the treatment of this common and chronic disease.

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