

Experimental models in asthma

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Summary

Histamine is an important mediator released from activated mast cells in acute bronchoconstriction provoked by allergen, exercise, hypertonic stimuli and inhaled adenosine. Histamine may also contribute to the allergen-induced late asthmatic response probably following the recruitment and activation of basophils. H_1 -receptor antagonists partially attenuate these responses but greater inhibition when these drugs are combined with cysteinyl LT_1 -receptor antagonists indicative of an interaction between mast cell-derived mediators. The combination of two or more selective mediator antagonists in the treatment of allergic disorders such as asthma offers a new therapeutic approach worthy of careful appraisal.

Keywords: allergen, exercise, hypertonic stimuli, inhaled adenosine, basophils

The role of histamine as a mediator of asthma

The recognition by Dale and Laidlaw [1] that histamine could be released from anaphylactically challenged lung tissue provided the basis for what was to be the beginning of the relentless search for mediators that could account for the airway dysfunction characteristic of asthma. The existence of abundant mast cells and basophils in the airway wall of patients with asthma [2], together with the many pharmacological actions mediated through H_1 receptors that histamine is able to produce pertinent to the pathophysiology of asthma, including smooth muscle contraction, increasing microvascular permeability, stimulating both local and central neural reflexes and augmenting mucous secretion provides the biological basis for why histamine should be considered as a mediator of asthma. Indeed, inhalation of increasing doses of histamine has long been used as a provocation test to aid in the diagnosis of asthma by revealing increased airways responsiveness [3].

However, it is the availability of selective receptor antagonists for the various histamine receptors that have enabled the contribution of this mediator to be placed into context of the many mediators that contribute to the asthmatic response, both alone and in combination with other autacoids. When discussing the potential role of histamine as a mediator in asthma, it is inevitable that one

must turn to the role of mast cells and basophils in this disease.

The mast cell as a mediator-secreting cell in asthma

Although mast cells are found throughout the human body and especially at mucosal surfaces, the airways seem to be a particularly rich source of this cell type [4]. While various postmortem and biopsy studies have suggested that mast cell numbers increase in the airways of patients with asthma, this is usually restricted to those with more severe disease. However, in active asthma and following exposure to allergen, mast cells have been shown to migrate from the submucosa into the epithelium where they are more likely to come into contact with inhaled allergen and other noxious stimuli. The majority of mast cells present in the epithelium and submucosa of normal and asthmatic airways belong to the tryptase-only type [5] that originate from $CD34^+$ precursor cells in the presence of both stem cell factor and mast cell growth factors derived from activated T lymphocytes of the Th2 subtype [6]. The bronchial epithelium and adjacent fibroblast and myofibroblasts provide an ample source of stem cell factor while T cells belonging to the Th2 phenotype are sources of cytokines that are considered important in polarizing the mast cell type towards the MC_T cell, namely interleukins-3, -4 and -9. In various combinations these cytokines are also involved in the regulation of the expression of the high affinity IgE receptor ($FC_\epsilon R1$), but recently it has also been shown that the level of expression

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of this receptor is intimately related to the level of circulating IgE [7]. There have been many studies now documenting the presence of elevated levels of histamine in bronchoalveolar lavage fluid obtained from patients with active asthma [8,9], indicating that these cells are in an activated state in this disease. Elevated histamine concentrations in lavage fluid are accompanied by other mast cell dependent mediators, including prostaglandin D₂ [10], leukotriene C₄ [11] and the protease, tryptase [9,12]. It is highly unlikely that histamine released in this way will not be acting alone, but in combination with the pharmacological and enzymic effects of the other mast cell-derived products.

The airway mast cell is also an important source of cytokines, including IL-3, IL-4, IL-5, IL-6, IL-8 (and other chemokines) and TNF- α , which are not only stored preformed within the secretory granules of these cells but upon dimerization of Fc ϵ R1 are newly generated through transcription mechanisms [13–15]. Once activated, human airway mast cells continue to generate and release cytokines, such as IL-5 and TNF- α , for up to 48–72 h [15]. It is therefore clear that the signalling mechanisms involved in the explosive release of mast cell mediators are separate from that involved in the generation and release of newly formed cytokines.

In order to explore the contribution of histamine as well as other mediators to the asthmatic response, it is important to examine its contribution to known factors that aggravate the asthmatic airway.

The role of histamine in allergen-induced bronchoconstriction

It has long been recognized that inhalation of allergen extracts to which the asthmatic patient is specifically sensitized leads to an early and late phase of airway obstruction accompanied by a prolonged increase in bronchial hyperresponsiveness [16]. The early asthmatic reaction is accompanied by a pulse of histamine or its metabolite N γ -methyl histamine in bronchoalveolar lavage, peripheral blood and in the urine [9,16–18]. Pre-treatment of asthmatic patients with a range of different H₁-receptor antagonists with varying potency at the H₁ receptor have provided overwhelming evidence for the importance of histamine in mediating the first phase (0–7 min) of the early asthmatic reaction [19–21], whereas prostaglandin D₂ [22] and the cysteinyl leukotrienes [23], both of which have to be newly generated before release, contribute most to the bronchoconstriction observed 7–20 min following allergen exposure.

There is still some confusion as to the potential role of histamine in the late asthmatic reaction as different antihistamines appear either not to inhibit or to partially inhibit

this response [24–26], without affecting non-specific bronchial hyperresponsiveness. It has been argued that during the late asthmatic reaction, there is an influx of basophils and that these cells, when activated, contribute, at least in part, to the bronchoconstriction observed between 6 h and 24 h after allergen exposure [27]. However, as up to 80% of the late asthmatic response can be inhibited by antagonizing the cysteinyl LT₁ receptor with such agents as montelukast and zafirlukast, an important role for histamine in this response is rather undermined [23,28]. However, in patients with only mild asthma who can tolerate a high dose of inhaled allergen, basophil influx and release of histamine from these cells becomes increasingly important. The availability of monoclonal antibodies that can now be used to identify basophils through the presence of specific proteins within their granules [29,30] will provide an opportunity for defining once and for all the role of these cells in this response and in asthma more generally.

Exercise- and cold air-induced asthma

A second important stimulus for asthma, in which histamine plays a key role, is the acute bronchoconstriction brought on by exercise [31]. Although there has been extensive debate about the mechanisms of exercise-induced asthma, it is now generally thought that the acute bronchoconstriction following a burst of exercise in patients with asthma is the consequence of increased activation of mast cells on the surface of the airways with release of histamine, prostaglandin D₂ and leukotrienes. While it has been very difficult to detect increased levels of mast cell mediators in bronchoalveolar lavage following either exercise or a related stimulus [32], it seems most likely that this is due to transient activation of mast cells restricted to the surface of the airways and not deeper in the airway wall. Recently Reiss *et al.* have demonstrated increased levels of the LTC₄ metabolite LTE₄ in the urine of asthmatic patients shortly after experiencing exercise-induced bronchoconstriction [33]. Moreover, prior treatment of asthmatic patients with selective H₁-receptor antagonists results in attenuation of the bronchoconstrictor response amounting to 30–50% [34, 35]. The remainder of the bronchoconstriction appears to be due to the release of cysteinyl leukotrienes as receptor antagonists of these mediators remove a further 60–70% of the exercise-induced bronchoconstriction [33,36,37]. Thus, together, a combination of an antihistamine with a cysteinyl LT₁ antagonist should be very effective in blocking exercise-induced asthma although clinical trials have yet to be undertaken to demonstrate this.

The mechanisms whereby exercise activates mast cells in the asthmatic airway is thought to be due to the inability of the bronchial epithelium to adequately condition the increased volume of respired air associated with exercise

to full humidity [38]. The precise reasons for this are not known but it has been suggested that there may be a primary defect of the bronchial epithelial cells in being able to adequately transport chloride ions and water to the airway surface in asthma, thereby resulting in a hyperosmolar environment [31]. Cytokine primed mast cells are extremely sensitive to hyperosmolar stimuli [39] and this property has recently been utilized as a method of measuring indirect bronchial hyperresponsiveness [40]. Thus, it is not surprising that H_1 -receptor antagonists are highly effective at inhibiting bronchoconstriction caused both by isocapnic hyperventilation and by inhalation of hypertonic saline but, again, the effectiveness of a combination of H_1 - and cysteinyl LT_1 -receptor antagonists in blocking these responses has yet to be evaluated.

Adenosine-induced bronchoconstriction

In 1983 we first described the bronchoconstrictor effect of inhaled adenosine and its 5'-nucleotides (AMP, ADP) in patients with asthma [41]. Over the following 10 years, we provided evidence that this bronchoconstriction results from the selective activation of airway mast cells with the release of preformed mediators, including histamine [42]. The precise mechanism probably involves activation of the A_{2b} receptor which, in some way, is linked to the amplification of mast cell degranulation [43]. Mast cells obtained from the airway surface of patients with asthma are hyperresponsive to adenosine and its analogues and A_{2b} receptor antagonist, such as enprofylline, have the capacity to inhibit bronchoconstriction produced by inhaled adenosine or AMP.

That most of the bronchoconstriction provided by this stimulus is histamine-mediated has derived from studies demonstrating the release of histamine into the blood and airway lining fluid following inhalation [44] or local challenge with AMP [45] and also the almost complete inhibition of bronchoconstriction provided by the prior administration of selective H_1 -receptor antagonists [46]. The factors that control the functional activity of the A_{2b} receptor on airway mast cells is not known nor the importance of this pathway in reducing the threshold which mast cells will respond to other stimuli including allergen. As mast cells express both A_{2b} receptors linked to adenylate cyclase via G_s protein as well as to PI-specific phospholipase via CG_q [43], it is possible that the reduced efficiency of G_s coupling, as occurs in asthma in association with impaired β_2 -adrenoceptor responses [47], leaves the G_q -PLC transduction mechanism linked to mast cell degranulation unopposed.

The role of histamine in nocturnal asthma

Nocturnal asthma is regarded as one of the principal markers of asthma severity. Although its precise mechanisms have yet

to be fully elucidated, we have recently shown that a large nocturnal decrease in pulmonary function is associated with an influx of activated T cells, eosinophils and mast cells [48]. The precise sequence of events has yet to be clarified but our recent work would suggest the selective upregulation of eosinophilopoietic cytokines, including IL-5 in mast cells and possibly T lymphocytes present in the most peripheral airways and alveoli. The capacity of cysteinyl LT_1 -receptor antagonists to effectively reduce nocturnal asthma provides additional evidence for the mediator basis of this component of the disease [49]. While antihistamines alone have relatively little efficacy in this form of asthma, it would certainly be worthwhile to further investigate the combination of antihistamine and leukotriene modifying drugs particularly in more severe disease.

Interaction of histamine with other mediators in the pathophysiology of asthma

Up until recently, when considering the origin and progression of asthma, individual mediators of inflammation have largely been considered in isolation. However, in real life this would seem to be highly unlikely. The study of how mediators interact in asthma is only just beginning to be studied. For example, it is known that histamine is a powerful stimulus for the transport of P-selectin from the Weibel Palisade bodies in the microvascular endothelium to the cell surface [50]. This adhesion molecule plays an important function as a lectin by interacting with its carbohydrate counter ligand expressed on migrating leucocytes, including neutrophils, eosinophils and T cell, to induce rolling and margination. The cytokine induced expression of E-selectin and ICAM-1 (TNF- α , IL-1 β and interferon- γ) and VCAM-1 (TNF- α , IL-4 and IL-13) provides the secondary stimuli which again may originate from the mast cell or other cells leading to tight binding of the leucocyte to the endothelium and transendothelial migration, a key feature observed in the late asthmatic response [51].

There may be more direct ways in which histamine may interact with other mediators of asthma, although simple studies of inhaling combinations of histamine and PGD_2 and histamine and LTD_4 have not yet revealed this. Nevertheless, the powerful influence of combining an H_1 antagonist with a cysteinyl LT_1 receptor antagonist in allergen-induced early and late asthmatic reactions provides a rational basis for developing a new type of therapeutic strategy for managing patients with asthma [52]. Indeed, a recent clinical trial in which the antihistamine, Loratidine, has been combined with the cysteinyl LT_1 receptor antagonist, Montelukast (10 mg of each daily), has shown efficacy in clinical asthma equivalent to that observed with a moderate dose of inhaled corticosteroid. The fact that neither antihistamine nor antileukotrienes are corticosteroids and can

both be taken orally once daily, provides an attractive alternative for asthma management, particularly in those patients in whom compliance with inhaled controller drugs is low.

Concluding comments

As a pharmacological mediator of asthma, there can be little doubt that histamine also contributes to the clinical expression of this disease. In most cases of asthma it seems most unlikely that antihistamines alone will offer much in the way of clinical benefit [53]. However, these drugs, when combined with other mediator antagonists or inhibitors such as leukotriene modifiers and cyclooxygenase-2 inhibitors (to reduce PGD₂ and thromboxane A₂ production) is beginning to reveal a new approach to treat asthma that deserves careful and rigorous evaluation.

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