

Mechanisms and significance of adenosine-induced bronchoconstriction in asthma

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The recognition that inflammatory processes play an important role in the pathogenesis of bronchoconstriction in asthma has led to a relentless search for individual chemical substances which may mediate disordered airway function. There is now compelling evidence that the immediate bronchoconstrictor response following provocation of the airways of atopic asthmatic subjects with inhaled allergen occurs through the release of bronchospastic mediators from mast cells activated on the surface of the bronchial mucosa (14). However, these cells and their mediators, which may be responsible for pathogenesis of the allergen-induced late asthmatic reaction and, of more importance, the induction of increased non-specific airways reactivity, remain to be identified.

It is well recognised that allergen challenge of isolated human airways or lung fragments releases both primary and secondary mediators of inflammation from a variety of cells. Adenosine represents one such inflammatory product which is released from a wide variety of cells following both immunological and non-specific stimulation (13), and this mediator may therefore contribute to the inflammatory reaction in asthmatic airways. Adenosine is a ubiquitous naturally occurring purine nucleoside that is released from a wide variety of cells under conditions of stress, e.g. hypoxia or cell activation under conditions of energy deficit (22). Most of the adenosine found extracellularly is generated from the 5'-nucleotidase cleavage of adenosine 5'-monophosphate (AMP) (2). Once released from an activated cell, adenosine may either be taken back up into neighbouring cells

where it is metabolised back to adenine nucleotides or degraded to inosine, hypoxanthine and uric acid. Alternatively, extracellular adenosine may serve as an autacoid by stimulating specific cell surface receptors which either stimulate (A_2) or inhibit (A_1) adenylate cyclase linked to the formation of intracellular 3'5'-cyclic adenosine monophosphate (cyclic AMP) (16). Synthetic analogues of adenosine, such as N-ethylcarboxamide-adenosine and L-N⁶-phenylisopropyl adenosine, exhibit some specificity for stimulating A_2 and A_1 receptors respectively. In addition, methylxanthines such as theophylline and 8-phenyltheophylline, are competitive antagonists for adenosine at its cell surface purinoreceptors (4).

A role for adenosine as a potential bronchoconstrictor mediator in asthma was first suggested by the demonstration that increasing inhaled concentrations of the purine nucleoside provoked bronchoconstriction in patients with asthma, whereas the related nucleosides, guanosine and inosine, had no effect (9, 19). Over the same concentration range, inhaled adenosine was shown to have no discernible effect on airway calibre of non-asthmatic airways. Both inhaled AMP and adenosine 2'5'-diphosphate also provoked bronchoconstriction in asthma in similar molar concentrations to adenosine (19). *In vivo* and *in vitro* studies in man and animals have shown that adenine nucleotides are rapidly hydrolysed to adenosine (3, 26), suggesting that the bronchoconstrictor activity of adenine nucleotides in asthma probably occurs after their *in vivo* conversion to adenosine.

Interest in the potential role of adenosine as a bronchoconstrictor mediator in asthma was heightened when it was demonstrated that theophylline administered either by inhalation (10) or orally (18) preferentially inhibited bronchoconstriction provoked by adenosine when compared to that produced by histamine, and raised the possibility that adenosine antagonism may in part account for some of the therapeutic effects of methylxanthines in asthma. At first sight this seemed an attractive hypothesis, particularly since the accepted therapeutic concentrations of theophylline in plasma (10–20 µg/ml) corresponded with those concentrations which effectively inhibit the purinoceptor effects of adenosine and were some 10 times lower than that required to effectively inhibit cyclic AMP phosphodiesterase (12).

Structure activity studies with a variety of substituted xanthines in animals indicated that adenosine antagonism could not account for the bronchodilator effect of this group of drugs (23), but could account for a wide variety of side effects such as CNS (11) and cardiac (27) stimulation. One compound emerging from this work was enprofylline which was reported to be approximately 3–5 times more potent than theophylline as a bronchodilator and yet be free of adenosine antagonistic activity (23).

To clarify the role of adenosine antagonism as a mechanism for methylxanthine's bronchodilator action in asthma, we have compared the relative protective effects of enprofylline and aminophylline on histamine and AMP-induced bronchoconstriction in seven patients with atopic asthma (6). Both drugs were administered as intravenous infusions to achieve steady state plasma concentrations and produce approximately equivalent bronchodilatation. Intravenous saline was used as placebo in this double-blind study. On separate days during each of the three infusions, bronchial provocation was undertaken with increasing doubling concentrations of inhaled AMP, histamine or repeated inhalations of saline, and airways calibre followed as changes in forced expiratory volume in 1 s (FEV₁). Both aminophylline and enprofylline produced an equivalent protection of the airways against inhaled histamine. How-

ever, theophylline was 4–5 times more effective than enprofylline in protecting the airways against the bronchoconstrictor effect of inhaled AMP, suggesting a specific form of antagonism beyond functional antagonism. We conclude from this study that adenosine antagonism is most unlikely to account for the bronchodilator activity of methylxanthines in asthma, but cannot totally exclude that such a pharmacological action could contribute to other beneficial effects of these drugs in this complex disease.

Investigation of the effect of methylxanthines on adenosine- and AMP-induced bronchoconstriction strongly suggests that the constrictor effects of these agents on asthmatic airways occurs through stimulation of specific cell surface purinoreceptors. In support of this suggestion is the demonstration that the adenosine uptake blocker, dipyrindamole has a preferential potentiating effect on bronchoconstriction provoked by adenosine when compared to that provoked by histamine (8). These studies, however, provide no information about which cells may be involved in mediating adenosine's effect in asthma. Further investigation showed that responsiveness of the airways to both adenosine and AMP was only weakly correlated with indices of non-specific bronchial responsiveness as determined by methacholine or histamine bronchial provocation (5, 8). As observed with allergen provocation, it is most likely that activation of an interceding cell or cells mediates the bronchoconstrictor activity of adenosine, possibly through the elaboration of inflammatory mediators. In support of this hypothesis is the demonstration that the mast cell stabilising drugs, sodium cromoglycate and nedocromil sodium, are powerful inhibitors of adenosine and AMP-provoked bronchoconstriction (1, 7).

In 1978 Marquardt et al. (20) reported that adenosine and synthetic adenosine analogues could potentiate calcium ionophore-stimulated mediator release from rat peritoneal mast cells. Studies on human lung mast cells (15) and mouse cultured IL-3-dependent mast cells (21) confirmed this observation and suggested a mechanism involving the activation of cell surface A₂-purinoreceptors. Marquardt et al. (21) have further shown that adenosine and A₂-selec-

tive synthetic analogues have a far greater effect in augmenting the release of granule-derived preformed mediators from activated mouse mast cells than newly generated mediators. This mechanism provided an attractive explanation for bronchoconstriction provoked by adenosine in asthma. In support of this suggestion we have shown that AMP, albeit at fairly high concentrations, provoked bronchoconstriction in non-asthmatic subjects only if they were atopic, suggesting the possibility of a mast cell or IgE-dependent mechanism for the response (5). Definitive evidence for mast cell involvement in adenosine and AMP-provoked bronchoconstriction has been obtained by showing that terfenadine and astemizole, two potent and selective H_1 -receptor antagonists, inhibit AMP-induced bronchoconstriction by over 80% (25).

Our current working hypothesis on the mechanism of action of adenosine as a bronchoconstrictor substance when inhaled by atopic subjects, is that the nucleoside accentuates release of spasmogenic mediators from preactivated mast cells on the bronchial mucosa leading to bronchoconstriction. It remains possible that some of the bronchoconstrictor response with adenine derivatives involves stimulation of bronchoconstrictor reflexes. However, we have been unable to demonstrate any protective effect of a highly potent muscarinic cholinergic antagonist, ipratropium bromide (1 mg inhaled) on the airway effects of inhaled adenosine (17).

In the initial studies describing the bronchoconstrictor effect of inhaled adenosine, both atopic and non-atopic asthmatic subjects responded in a similar manner. Since selective antagonism of H_1 receptors in the airways of patients with intrinsic asthma also has a major inhibitory effect on AMP-induced bronchoconstriction (24), it is probable that a similar mast cell-dependent mechanism accounts for the constrictor airways response as suggested for atopic asthma. If this interpretation is correct, then AMP hyper-responsiveness of the airways in the presence of intrinsic asthma implies that mast cells in the bronchial mucosa of these patients are preactivated, although probably not by an IgE-dependent mechanism.

The recent demonstration that increased cir-

culating concentrations of adenosine occur with bronchial provocation with either inhaled allergen or methacholine suggests that this purine nucleoside is released during an asthmatic response (19). The precise cellular origin of the nucleoside and its significance in promoting bronchoconstriction with inhaled allergen is at present not known. However, studies involving this nucleoside have raised some interesting possibilities as to the role of mast cells in the pathogenesis of airflow obstruction in various forms of asthma which will hopefully stimulate further research into the contribution of inflammatory mediators in the pathogenesis of asthma.

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