ADENOSINE STIMULATES REFLEX CHOLINERGIC MEDIATED CONTRACTIONS OF TRACHEAL SMOOTH MUSCLE *IN SITU*

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Introduction: Adenosine has been shown to induce bronchospasm in asthmatic but not healthy subjects (Cushley et al.,1985) Mast cell derived mediators are largely implicated in the airways response to adenosine. However the role of a neuronal mechanisms is implicated since bronchoconstriction to AMP (adenosine 5'monophosphate) was attenuated by ipratropium bromide (Polosa *et al.,* 1991). In this current study we have sought to investigate whether N^6 – cyclo-pentyl adenosine (CPA), which is an A_1 receptor agonist can initiate reflex cholinergic contraction of tracheal smooth muscle *in situ* in guinea pigs.

Methods: Male Dunkin-Harley guinea pigs (300-500g) were anaesthetised (urethane 1.5g/kg ip.) connected to a ventilator (4ml/kg; 60 breaths/min) then paralysed (succinylcholine 2mg/kg subcutaneously) and prepared for the measurement of tracheal tension *in situ,* as described elsewhere (Mazzone *et al*., 2002). Changes in tracheal tension and airway mechanics were measured simultaneously in response to CPA, histamine and bradykinin. The abdominal vena cava was cannulated for intravenous administration of drugs whilst the abdominal aorta was cannulated for the measurement of blood pressure. Increase in airways resistance (R_L) and decrease in compliance (C_{dyn}) were also measured in response to intravenously administered drug. Cholinergic tone was reversed by adding 1μM atropine to the tracheal perfusate. DPCPX (A1 receptor antagonist), pyrilamine) (H1 receptor antagonist) and WIN64338 (1 μM) (B2 receptor antagonist) were also added to the tracheal perfusate to rule out any direct action of CPA, histamine and bradykinin, respectively on tracheal tension.

Results: Changes in R_L and C_{dyn} are expressed as a percentage increase or decrease respectively from baseline.Results are expressed as mean ± s.e.m, n=5-9 CPA, histamine and bradykinin dose response curves (1-4μg/kg) IV were performed. CPA and bradykinin induced an almost negligible increase in airways R_L (CPA: 2μg/kg; 7.2±1.1%), (BK: 2μg/kg; 12.8±3.6) and (C_{dyn}) (CPA: 2μg/kg; 3.9±1.8) (BK: 2μg/kg; 9.8±2.4). In contrast, histamine induce a significant bronchoconstrictor response (R_L: 2μg/kg; 55.1±14.0) (C_{dyn}: 2μg/kg; 36.7±11.0). Concomitant with these changes in airway lung mechanics, CPA, histamine and bradykinin also contracted guinea-pig trachea (grams tension) *in situ* (CPA:2μg/kg; 0.07±0.02g; histamine:2μg/kg; 0.16±0.02g and bradykinin:2μg/kg; 0.092±0.03g). In all experiments atropine blocked the reflex to CPA, bradykinin and histamine confirming activation of reflex cholinergic pathways.

Conclusion: CPA does not cause bronchospasm in normal animals but still elicited a cholinergic reflex, which is indicated by tracheal contraction. Further experimentation is underway to define the mechanisms by which the cholinergic reflex to CPA is initiated.

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