Inhibition of inhaled metabisulphite-induced bronchoconstriction by inhaled frusemide and ipratropium bromide

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- 1 The effect of inhaled frusemide and high dose inhaled ipratropium bromide on bronchoconstriction induced by inhaled metabisulphite was studied in 10 atopic volunteers.
- 2 Frusemide (40 mg), ipratropium bromide (0.5 mg) or saline placebo were administered by nebuliser in a double-blind fashion, prior to construction of a dose-response curve to metabisulphite (2.5–100 mg ml⁻¹).
- 3 Geometric mean of the provocative dose of metabisulphite that caused a 35% fall in specific airways resistance (sGaw) after placebo was 13 (95% confidence intervals CI 4–36 µmol) compared with 36 (16–78) µmol after ipratropium bromide and 45 (22–94) µmol after frusemide.
- 4 Mean maximum fall in sGaw was 49 (40–57)% after placebo, 11 (0–22)% after frusemide and –1 (–25–22)% after ipratropium bromide.
- 5 Frusemide significantly protected against metabisulphite induced bronchoconstriction ($P < 0.005$). The protection from high dose ipratropium bromide was also significant ($P < 0.05$), but the response was more variable between subjects.

Keywords sodium metabisulphite bronchoconstriction frusemide ipratropium bromide

Introduction

Metabisulphites are widely used as food preservatives but their ingestion can cause severe bronchoconstriction and other systemic effects in sensitive individuals (Baker *et al.*, 1982). Inhaled sodium metabisulphite (MBS) induces bronchoconstriction, in a dose-response fashion, in both atopic non-asthmatic and in asthmatic subjects (Dixon & Ind, 1988; Nichol *et al.*, 1990). MBS bronchoconstriction is blocked by inhaled nedocromil sodium and sodium cromoglycate (Dixon & Ind, 1990; Wright *et al.*, 1990). We found partial inhibition by the anticholinergic agent oxitropium but others found no effect of ipratropium bromide 200 µg (Nichol *et al.*, 1989). We measured specific airways conduction (sGaw) whereas the other study determined FEV₁. Nebulised MBS releases high concentrations of sulphur dioxide (SO₂) and it is likely that bronchoconstriction is due to SO₂ though the exact mechanism is unclear (Dixon & Ind, 1988). Previous studies in animals (Grunstein *et al.*, 1977) and man (Snashall & Baldwin, 1982; Tan *et al.*,

1982) have shown that SO₂ induced bronchoconstriction is inhibited by anticholinergic blockade. Inhaled frusemide has been shown to block bronchoconstriction induced by exercise (Bianco *et al.*, 1988), fog (Robuschi *et al.*, 1989), adenosine (Polosa *et al.*, 1990) and allergen (Bianco *et al.*, 1989) but not bronchoconstriction after histamine (Robuschi *et al.*, 1990) or methacholine (Nichol *et al.*, 1990). Recently inhaled frusemide has been reported to block MBS induced bronchoconstriction (Nichol *et al.*, 1990).

It has been suggested that frusemide may act on airway nerves (Ventresca *et al.*, 1990) though it is not clear whether this is on afferent or efferent pathways.

The aim of this study was to examine the effect of pretreatment with ipratropium bromide (IB) in a high dose which would be expected to block cholinergic nerve activity and to compare this effect with that of inhaled frusemide (F) on MBS induced bronchoconstriction in atopic subjects.

Methods

Subjects

Ten atopic volunteers were studied; five were female and five had mild asthma. Atopic status was determined by two or more positive skin prick tests to a panel of 12 common allergens. Asthma was defined according to the classification criteria of the American Thoracic Society (American Thoracic Society, 1962). Of the asthmatics, one subject used regular and four used occasional inhaled β_2 -adrenoceptor agonists. None was using inhaled steroids or other therapy. Mean age was 29 years (range 21–40). All subjects had previously shown a greater than 35% fall in sGaw in response to a prior MBS challenge. All gave written informed consent to the study, which had been approved by the Ethics Committee of Hammersmith Hospital.

Study design

Subjects attended on 3 study days. Specific airways conductance (sGaw) was determined by computerized body plethysmography (Chowienzyk *et al.*, 1981). Subjects were pretreated with either F 40 mg (10 mg ml^{-1}) (Hoechst, Hounslow, Middx), high dose IB 0.5 mg (250 $\mu\text{g ml}^{-1}$) (Boehringer Ingelheim, Bracknell, Berks) or saline (S) placebo in a randomized double-blind fashion. All drugs were made up to 4 ml with saline and delivered by Cirrus nebuliser (output 0.19 ml min^{-1}) over 10 min. After a further 10 min MBS challenge was carried out, MBS (Sigma, Poole, Dorset) being delivered by Mefar dosimeter (5 breaths, output 0.24 ml per breath) in increasing concentrations (2.5, 5, 10, 25, 50, 100 mg ml^{-1}) until a greater than 35% fall in sGaw was recorded. sGaw was measured twice before drug administration then 1 min after drug, saline and each concentration of MBS delivered during the MBS challenge.

Statistics

The PD_{35}MBS (cumulative provocative dose of MBS causing a 35% fall in sGaw) was calculated by linear interpolation of the log dose-response curve. Logarithmically transformed PD_{35} values were analysed by multifactorial analysis of variance to assess the effect of pretreatment with F and IB on MBS. PD_{35} values were expressed as geometric mean (GM PD_{35}) with 95% confidence intervals (CI). In cases where a 35% fall in sGaw was not achieved with 100 mg ml^{-1} MBS a value of 120 μmol was allocated.

Results

All subjects coughed at concentrations of MBS greater than 50 mg ml^{-1} , irrespective of bronchoconstriction. The procedures were well tolerated and no ill effects were observed. Baseline sGaw did not differ significantly on the 3 study days and it did not alter significantly after F or S. After IB there was significant bronchodilation, mean sGaw increasing from 1.78 to 2.26 $\text{kPa}^{-1} \text{s}^{-1}$ ($P < 0.05$) (Figure 1). PD_{35}MBS for the 10 subjects for F,

Figure 1 Percent change in baseline sGaw 10 min after nebulised frusemide (F, 40 mg), ipratropium bromide (IB, 0.5 mg) and saline (S) control.

Figure 2 Log of geometric mean cumulative PD_{35}MBS in μmol and confidence intervals (vertical lines) after frusemide (F, 40 mg), ipratropium bromide (IB, 0.5 mg) and saline (S) control.

IB, and S are shown in Table 1 with corresponding geometric means and confidence intervals. GM PD_{35}MBS after frusemide was 45 (22–94) μmol compared with GM PD_{35}MBS of 13 (4–36) μmol after placebo. Frusemide significantly inhibited MBS induced bronchoconstriction compared with saline ($P < 0.005$) (Figure 2).

GM PD_{35}MBS after ipratropium bromide was 36 (16–78) μmol . Hence IB also significantly inhibited MBS induced bronchoconstriction compared with saline ($P < 0.05$). The effect of ipratropium bromide on MBS induced bronchoconstriction was variable; in seven subjects PD_{35} increased by > 2 fold while in the remaining three it either fell or did not change, (Table 1). There was no correlation between bronchodilatation produced by IB and protection against MBS-induced bronchoconstriction. Protection from ipratropium bromide did not differ significantly from that of frusemide.

The % fall in sGaw at the highest concentration of MBS inhaled on the placebo day was compared for the three groups. After placebo the subjects had a mean fall in sGaw of 49 (40–57)% compared with a fall of 11 (0–22)% with frusemide ($P < 0.001$) and -1 (-25–22)%

Table 1 Cumulative PD₃₅ metabisulphite (μ mol) for individual subjects

Subject	Frusemide	Ipratropium bromide	Saline
1	46	20	30
2	120	120	120
3	120	120	34
4	120	120	40
5	8	15	4
6	53	8	30
7	37	36	2
8	120	120	9
9	10	9	1
10	25	21	9
Geometric mean	45 ($P < 0.005$)	36 ($P < 0.05$)	13
Confidence intervals	22–94	16–78	4–36

with ipratropium bromide ($P < 0.01$). The correlation between protection by frusemide and IB when PD₃₅ values were compared was invalidated by four pairs of censored values. When % change in sGaw at the highest MBS concentration inhaled on placebo day was compared for frusemide and IB there was no significant correlation ($r = 0.63$).

Discussion

Frusemide significantly inhibited bronchoconstriction induced by inhaled metabisulphite. We demonstrated a mean 5.5 fold increase in PD₃₅MBS after frusemide compared with saline control. This confirms the results of Nichol *et al.* (1990) who showed a mean 3 fold protection. We administered an estimated dose of 24 mg of frusemide, calculated from pre and post nebulisation weights. This compares with their dose of 30 mg (Nichol *et al.*, 1990). Hence frusemide resembles nedocromil and sodium cromoglycate in blocking metabisulphite as well as other indirect bronchoconstrictor stimuli including exercise (Bianco *et al.*, 1988), fog (Robuschi *et al.*, 1989), adenosine (Polosa *et al.*, 1990), and allergen (Bianco *et al.*, 1989). Bronchoconstrictor challenges which act directly on bronchial smooth muscle such as inhaled histamine or methacholine are not inhibited (Nichol *et al.*, 1990). The mechanism of action of frusemide is still unknown though mediator cells or nerves rather than bronchial smooth muscles may be involved (Knox & Ajao, 1990).

We also found significant inhibition of MBS induced

bronchoconstriction by ipratropium bromide 0.5 mg, examining PD₃₅ or % fall in sGaw at the highest dose of MBS inhaled on placebo day. In a previous study of atopic non-asthmatic and mild atopic asthmatics we found that oxitropium bromide partially, but significantly, inhibited MBS induced bronchoconstriction (Dixon & Ind, 1988). However, Nichol *et al.* (1989) did not confirm these findings using ipratropium bromide 200 μ g in asthmatic subjects with more marked methacholine responsiveness. The other methodological differences between the two studies were that we induced less marked bronchoconstriction and this was measured by sGaw whilst Nichol *et al.* (1989) measured FEV₁. The present study confirms our earlier findings with significant inhibition of MBS induced bronchoconstriction by ipratropium bromide, albeit administering a large dose. This dose should produce maximum anticholinergic receptor blockade, however we also found a rather variable response between different subjects (Table 1). Seale *et al.* (1988) found partial protection of MBS-induced bronchoconstriction in three out of nine asthmatics by inhaled ipratropium bromide 500–700 μ g. In fact a variable inhibitory response from anticholinergic agents is not unusual in studies of induced bronchoconstriction. It is more common to detect a positive result if relatively less bronchoconstriction is induced and a selective measure of airway calibre such as sGaw is used (Gross & Skorodin, 1984). If MBS bronchoconstriction does depend on SO₂ release then blockade by anticholinergic agents would be anticipated both from animal experiments (Grunstein *et al.*, 1977; Nadel *et al.*, 1965) and from human studies (Snashall & Baldwin, 1982).

In summary, we have confirmed the observations of Nichol *et al.* (1990) that frusemide inhibits MBS induced bronchoconstriction as for other indirect challenges which are also blocked by cromoglycate and nedocromil. We have also confirmed our earlier finding that anticholinergic agents produce partial blockade although this varies from subject to subject. These results are compatible with the hypothesis that MBS stimulates sensory nerves producing cough and leading to bronchoconstriction by non-cholinergic and cholinergic pathways. Ipratropium bromide acts on the efferent parasympathetic limb as a competitive antagonist at post synaptic muscarinic receptors, and at this dose is probably producing near maximal achievable cholinergic blockade. Frusemide is more effective than ipratropium bromide in inhibiting MBS-induced bronchoconstriction suggesting a different site of action. It is possible that frusemide acts on an afferent limb by analogy with its inhibitory effect on capsaicin or low chloride ion induced cough (Ventresca *et al.*, 1990) but further investigation is required.

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