Plasma concentrations and urinary excretion of histamine after inhalation and subcutaneous injection of histamine

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- 1 Increased histamine concentrations are found in the plasma and urine following allergen challenge in allergic subjects. This study compared a controlled challenge with clinically relevant doses of inhaled and injected histamine, as indicative of an allergic response, in an attempt to validate the use of urinary histamine or 1-methylhistamine measurements as an objective, non-invasive diagnostic test.
- 2 Inhalation of histamine produced peripheral vasodilation, increased heart rate, a fall in partial expiratory flow rate (pEFR) and blood pressure, 'tight chest' and cough. Subcutaneous injection produced vasodilation and headache but no change in heart rate or blood pressure.
- 3 Plasma histamine concentrations were similar in the two studies. Inhalation of increasing doses of histamine through a nebuliser (output 0.13 ml min^{-1}) resulted in an increase from a mean of 0.30 to 1.65 ng ml^{-1} , with return towards baseline within 20 min. Injection of 1 mg histamine s.c. produced an increase from 0.32 to 1.4 ng ml^{-1} within 5 min, remaining above 1 ng ml^{-1} for 30 min.
- 4 There was a significant increase of 15.2 ng mg^{-1} creatinine in urinary histamine concentration following the injection of histamine ($P = 0.04$) and an increase of 11.4 ng mg^{-1} creatinine when histamine was given by inhalation ($P = 0.18$). Histamine excretion rate increased by 108 ng min^{-1} ($P = 0.04$) after inhalation and by 37.2 ng min^{-1} ($P = 0.09$) after injection. Urinary 1-methylhistamine concentrations were significantly raised following both histamine inhalation ($+ 238 \text{ ng mg}^{-1}$ creatinine; $P = 0.013$) and injection ($+ 180 \text{ ng mg}^{-1}$ creatinine; $P = 0.03$). Its excretion rate was also raised following inhalation of histamine ($+ 1641 \text{ ng min}^{-1}$; $P = 0.02$).
- 5 Our findings support the use of urinary measurements of histamine and its metabolite to augment diary card diagnosis of 'allergic' reactions.

Keywords histamine plasma urine absorption excretion

Introduction

The pharmacological effects of histamine and its metabolic fate in man are well documented after administration by a number of routes (Douglas, 1985; Schayer, 1956). However, few observations have been made of the comparative effects of subcutaneously injected and inhaled histamine on clinical symptoms and the concentrations of histamine in blood and its metabolites in urine.

Histamine has been implicated as one of the major mediators of the vasoactive effects seen during anaphylactic and anaphylactoid reactions. Studies which have shown raised plasma histamine concentrations following antigen challenge (Howarth *et al.*, 1985; Ind

et al., 1983) have, however, required stringent blood handling procedures, sensitive and specific assays, and repeated venepuncture. The alternative of measuring urinary histamine and its primary metabolite 1-methylhistamine has received little attention.

Histamine is metabolised by two pathways. The major pathway involves conversion by histamine *N*-methyl transferase to 1-methylhistamine and finally to 1,4-methyl imidazole acetic acid. The other is via oxidative deamination by diamine oxidase (histaminase) and monoamine oxidases (Schayer & Karjala, 1956). Once absorbed, histamine is converted rapidly to inactive metabolites. For any given dose of histamine, 10–15%

is excreted as 1-methyl histamine and only 0.5–1.5% as unchanged histamine.

The aims of the study were, firstly, to measure concentrations of histamine in plasma following subcutaneous injection or inhalation of histamine at doses chosen to mimic the effects of an allergen challenge in allergic subjects; and secondly, to measure the urinary excretion of histamine and 1-methyl histamine under the same conditions. A significant change in the urine data would be a useful adjunct to subjective diary card assessment of outpatient challenge with foods, drugs or additives.

Methods

Subjects

Male, non-asthmatic volunteers (three atopic; three non-atopic) were selected on the basis of a relative insensitivity to histamine bronchial provocation tests. Thus, they tolerated inhalation of a 64 mg ml⁻¹ solution of histamine acid phosphate given by a Wright nebuliser (output 0.13 ml min⁻¹) without producing a 40% fall in pEFR₇₀*. The subjects were not on medication and had not had an upper respiratory tract infection within 2 weeks of the study. The mean age, height and weight of the volunteers was 35 years (range 27–48), 1.82 m (range 1.75–2.03) and 78 kg (range 53–90), respectively. Informed written consent was obtained in all cases and the protocol was approved by an independent Ethics Committee.

Dietary restrictions

The subjects were placed on a low histamine diet 72 h prior to and throughout the study. This excluded foods known to be high in histamine content, i.e. alcoholic and fermented products, cheese, yoghurt, smoked and tinned fish and meat, spinach and aubergines. The subjects were also instructed not to take strenuous exercise during the 72 h urine collection period.

Study design

The study was conducted over 3 consecutive days. Day 1 provided baseline measurements for urinary histamine and 1-methylhistamine from sequential 6 hourly collections of urine. On day 2, immediately after emptying the bladder, each subject underwent a single histamine bronchial provocation test with saline diluent followed by 2-fold increasing concentrations of histamine (4, 8, 16, 32 or 64 mg ml⁻¹) delivered by a Wright nebuliser (output 0.13 ml min⁻¹) until, in two consecutive manoeuvres, the percent fall in pEFR₇₀ from control exceeded 40%, or unacceptable signs and symptoms of systemic absorption of histamine were experienced, or the top dose of histamine was reached.

Venous blood samples were taken on two occasions

*pEFR₇₀ is the expiratory flow rate at a lung volume equivalent to 30% of the vital capacity above the residual volume, measured during a forced expiratory manoeuvre following a partial (i.e. sub maximal) inspiration.

prior to challenge and immediately after the end of each 2 min inhalation period, and also at 2, 5, 10 and 20 min after inhaling the top histamine dose.

Urine samples were collected as soon as possible following provocation (usually within 30–40 min with water diuresis). The sample time was based on the assumption of rapid excretion of histamine following an immediate anaphylactoid type response. Subjects then collected urine in 6 hourly aliquots for the remaining 24 h. Collection to 24 h was chosen to account for release of histamine following a delayed response or secondary histamine release at the target organ. On day 3, the same procedures were followed with the substitution of a single subcutaneous injection of 1 mg histamine for the bronchial provocation procedure. Blood samples were collected at 5 min intervals for 30 min.

Pulse rate, blood pressure and clinical symptoms were recorded throughout the experiment. Adverse events were elicited by a symptom checklist, before dosing and at the end of both the bronchial provocation test and subcutaneous injection. In addition, all spontaneously reported adverse events were recorded throughout the study. Symptom severity was graded by the volunteer on a 1–5 point scale. All pharmacodynamic measurements were assessed relative to baseline observations, as this was an open, non-randomised study. Plasma and urinary histamine and urinary 1-methylhistamine concentrations were measured blind.

Histamine bronchial provocation tests

Histamine acid phosphate at concentrations of 4, 8, 16, 32, 64 mg ml⁻¹ (salt) in sterile saline were prepared daily. Inhalation of doubling concentrations of histamine preceded by a saline control was carried out according to the method of Cockcroft *et al.* (1977) using a Wright nebuliser (delivery 0.13 ml min⁻¹ of solution) for 2 min at each dose, given 5 min apart. Lung function was assessed from partial expiratory flow rates.

Subcutaneous challenge

Histamine acid phosphate injection BP 1 mg (salt) in 1 ml (Martindale Pharmaceuticals Ltd, Romford, UK) was administered by subcutaneous injection in the deltoid region.

Blood sampling

Blood samples (10 ml) were withdrawn from an indwelling venous cannula, the first 2 ml being discarded, and collected into EDTA, mixed and put on ice. The samples were centrifuged within 15 min of collection at 4° C. The upper 50% of the plasma was removed and stored at –70° C.

Plasma histamine assay

Plasma histamine was measured using a single isotope radioenzymatic assay based upon the methods described by Guilloux *et al.* (1981) and Church *et al.* (1982). The assay was modified by the addition of 5 µl of diamine oxidase (DAO) (1 iu ml⁻¹) to 50 µl of each sample and these were assayed in parallel with non-digested samples

to correct for non-specific methylation. Standard curves were constructed from pooled frozen plasma samples spiked with 0, 0.05, 0.1, 0.2, 0.5, 1, 2, 5 and 10 ng ml⁻¹ histamine. The calibration curve was linear and passed through the origin. The limit of determination was 0.05 ng ml⁻¹. The intra- and interassay coefficients of variation were 8 and 11%, respectively ($n = 8$), for a 0.5 ng ml⁻¹ standard.

Urine collection

Urine samples were collected for 24 h periods prior to bronchial challenge, and on the days of bronchial challenge and subcutaneous challenge. Samples were pooled over successive 6 h periods and included the post-provocation urine sample. They were collected in polypropylene bottles containing 1 ml of 20% (w/v) chlorhexidine gluconate (ICI). Chlorhexidine at this concentration is known to inhibit the growth of histamine catabolising bacteria at 4° C. Urine concentrations were expressed as ng mg⁻¹ creatinine to correct for varying urine volume and urinary excretion rate was expressed in ng min⁻¹.

Urinary histamine assay

Histamine concentrations in the urine were measured according to the method of Myers *et al.* (1981). Briefly, urine samples were divided into two aliquots, centrifuged to remove particulate matter and duplicate measurements made on each aliquot. The histamine in one aliquot was degraded enzymatically by diamine oxidase (DAO). Both paired samples were then boiled, centrifuged and subjected to cationic exchange chromatography on DOWEX 50 W resin. Histamine was eluted, after washing with 0.1 M phosphate buffer and 1 M HCl, with 4 M HCl. The samples were neutralised with 10 M NaOH and assayed spectrofluorometrically (Siraganian & Brodsky, 1976). An internal standard of tritiated histamine was added prior to the extraction. The sample treated with diamine oxidase was a control for non-specific fluorescence, while the fluorescence measured in the other included that due to histamine. Therefore, the difference in fluorescence of the two samples was attributable to histamine. The limit of reliable assay was 1.5 ng ml⁻¹. The intra- and inter-assay coefficients of variation were 9 and 13%, respectively ($n = 8$), using a 25 ng ml⁻¹ standard.

Urinary 1-methylhistamine assay

1-Methylhistamine, the primary metabolite of histamine, was assayed using the method of Granerus & Wass (1984). Briefly, urine samples were extracted into salt-saturated alkaline butanol, back-extracted into acidified heptane, neutralised and applied to Cellex-P cationic exchange columns. Recovery and correction factors were determined using 3-methylhistamine as internal standard. The amines were eluted with 0.2 M borate buffer pH 8.5. The eluate was derivatised with *O*-phthaldehyde and assayed by reverse-phase-h.p.l.c., on a 15 × 4.6 mm 5 µm C18 ultrasphere column eluted with 0.1 M phosphate buffer containing a 0.4% v/v triethylamine (pH 6.4)-methanol-acetonitrile (3-2-1). The

limit of assay was 10.0 ng ml⁻¹. The intra- and inter-subject assay coefficients of variation were 7 and 9%, respectively ($n = 6$), using a 62.5 ng ml⁻¹ standard.

Creatinine assay

Creatinine was measured spectrophotometrically by the method of Jaffe (Wotton, 1986).

Statistical analysis

Unless otherwise stated results are given as means and standard errors of the mean (s.e. mean) or standard deviation (s.d.).

Urinary histamine concentrations and excretion rates following inhaled histamine (day 2) and subcutaneous histamine (day 3) were compared with the control results on day 1 by paired *t*-tests and 95% confidence intervals. The main comparison of interest was between the post-provocation sample and the 0–6 h control sample.

It should be noted that the power of the study to detect differences of clinical importance was low. Hence confidence intervals should be considered in association with *P* values.

Results

Plasma histamine following inhalation

The pooled estimate of the mean ($\pm$ s.e. mean) resting plasma histamine concentration in the two pre-dosing samples was 0.30 ± 0.06 ng ml⁻¹ (range 0.12–0.45 ng ml⁻¹). This rose with incremental histamine inhalation to a maximum concentration of 1.65 ± 0.17 ng ml⁻¹ ($n = 5$ subjects) at 2 min after the 64 mg ml⁻¹ inhalation. Concentrations fell below 1 ng ml⁻¹ at about 10 min after challenge and continued falling to 0.63 ± 0.25 ng ml⁻¹ by 20 min after challenge (Figure 1).

Figure 1 Group mean ($\pm$ s.e. mean) plasma histamine concentrations following histamine bronchial provocation tests in six subjects. Histamine was given in 2 min periods of tidal breathing at a concentration of 4–64 mg ml⁻¹ (sal = saline).

Plasma histamine following subcutaneous injection

Mean ($\pm$ s.e. mean) baseline plasma histamine concentrations were comparable with those on the previous day (before inhaled histamine) (0.32 ± 0.07 ng ml⁻¹; range 0.14–0.55 ng ml⁻¹). Mean concentrations rose immediately after injection reaching a maximum of 1.4 ± 0.5 ng ml⁻¹ at 5 min, and were above 1 ng ml⁻¹ for the duration of the study, indicating a more prolonged absorption of histamine from the subcutaneous site than from the lung (Figure 2).

The maximum circulating concentrations of histamine were similar following inhalation or subcutaneous injection of histamine.

Clinical symptoms

No subject experienced a fall in the forced expiratory volume in 1 s (FEV₁) of greater than 10%. One subject failed to reach the highest concentration of inhaled histamine solution (64 mg ml⁻¹) because of marked systemic symptoms occurring at an inhalation concentration of 32 mg ml⁻¹.

The pattern of clinical response varied substantially between the two challenge regimens. Histamine by inhalation produced flush in one subject at 16 mg ml⁻¹ and in all subjects at 64 mg ml⁻¹. Four subjects complained of cough at 32 and 64 mg ml⁻¹. Four complained of tight chest at the highest dose, but with a fall of less than 10% in FEV₁, compared with pre-challenge. However, there was a clinically significant fall (mean 45%) in partial expiratory flow rate at 70% maximum. In a previous study, these subjects were refractory to histamine at this dose.

Mean ($\pm$ s.d.) systolic and diastolic blood pressures pre-dosing were 127.0 ± 6.2 and 80.8 ± 7.9 mm Hg, respectively. Systolic blood pressure increased by 11.0 mm Hg (s.d. 6.0) to 137.6 mm Hg at 15 min after inhalation and diastolic blood pressure increased by 6.2 mm Hg (s.d. 8.8). Mean heart rate pre-dosing was 57.7 ± 12.1 beats min⁻¹, rising in proportion with incremental

Figure 2 Group mean ($\pm$ s.e. mean) plasma histamine concentrations following injection of 1 mg histamine subcutaneously in six normal subjects.

Figure 3 Mean ($\pm$ s.d.) cardiovascular and respiratory changes during and subsequent to a histamine bronchial provocation test (4–64 mg ml⁻¹) in normal non-asthmatic subjects ($n = 6$).

histamine inhalation to 90.4 ± 19.0 beats min⁻¹. On reaching the 64 mg ml⁻¹ dose level mean heart rate was significantly higher, with an increase of 33.4 (s.d. 10.5) beats min⁻¹ compared with its pre-dosing level. Heart rate returned to pre-dosing levels after 10 min (Figure 3). One subject's heart rate increased from 73 to 117 beats min⁻¹ and he experienced palpitations at the highest dose. His blood pressure dropped from 120/83 to 85/51 mm Hg.

Histamine given by subcutaneous injection resulted in flush appearing within 3 min in five of the six subjects and lasting for 9 min. There were no complaints of chest tightness and no subject coughed, although moderate to severe pulsatile headache was noted in three subjects within 3 min. This resolved within 10 min in all but one subject who experienced slight headache throughout the study. Nasal congestion was noted in one subject accompanying the pronounced flush. In contrast to the effect of inhaled histamine there were no substantial changes in either pEFR₇₀ or systolic or diastolic blood pressure.

Mean ($\pm$ s.d.) systolic and diastolic blood pressures pre-dosing were comparable with those on the previous day at 127.3 ± 11.8 and 79.3 ± 7.8 mm Hg, respectively. Systolic and diastolic blood pressure did not change following subcutaneous injection of histamine (Figure 4). Mean heart rate did not change significantly from its pre-dosing value of 65.0 ± 14.3 beats min⁻¹. However, one subject had a drop in blood pressure from 143/83

Figure 4 Mean ($\pm$ s.d.) cardiovascular and respiratory changes following subcutaneous injection of 1 mg histamine in normal non-asthmatic subjects ($n = 6$).

to 117/64 mm Hg, with a corresponding increase in heart rate from 67 to 97 beats min^{-1} after histamine injection.

Urinary histamine/1-methylhistamine concentration (Table 1)

Urinary histamine varied widely amongst subjects throughout the 3 study days. The mean ($\pm$ s.d.) urinary histamine concentration in the period 0–6 h on the control day 1 was 7.7 ± 5.1 ng mg^{-1} creatinine (range = 1.0–14.0 ng mg^{-1} creatinine). There was an increase of 11.4 ng mg^{-1} (95% CI: -6.1 , $+28.9$; $P = 0.18$) to 19.1 ng mg^{-1} creatinine on day 2 post-inhalation compared with the control day 0–6 h interval. Mean concentrations returned to values comparable with those obtained during the corresponding day 1 time intervals within the 0–6 h time interval and thereafter. Post-injection (day 3) urinary histamine was significantly higher with a mean difference of 15.2 ng mg^{-1} (95% CI: 0.8, 29.5; $P = 0.04$) to 22.9 ng mg^{-1} creatinine compared with that in the control day 0–6 h interval.

The mean ($\pm$ s.d.) urinary 1-methylhistamine concentration in the 0–6 h period on control day 1 was 99 ± 52 ng mg^{-1} creatinine (range = 40–194 ng mg^{-1} creatinine). There was a significant increase of 238 ng mg^{-1} (95% CI: 60.9, 414.8; $P = 0.013$) to 337 ng mg^{-1} creatinine on day 2 post inhalation. Values obtained in the 0–6 h post provocation period were raised by 126 ng mg^{-1} to 225 ng mg^{-1} creatinine compared with the corresponding control day values. Concentrations returned to values comparable with those on the control day after 12 h. Day 3 post injection concentrations were significantly higher, with a mean increase of 180 ng mg^{-1} (95% CI: 7.4, 352.6; $P = 0.03$) to 279 ng mg^{-1} creatinine compared with day 1 concentrations. Concentrations in the 0–6 and 6–12 h periods were higher than those observed on day 1.

Table 1 Group mean urinary histamine (Table 1a) and 1-methylhistamine (Table 1b) concentrations (ng mg^{-1} creatinine) over 24 h periods on the 3 study days. Day 1 control, day 2 histamine bronchial provocation test (4–64 mg ml^{-1}), day 3 subcutaneous histamine injection (1 mg). All mean differences are expressed relative to the control day

	Control day 1 Mean	Inhaled histamine day 2 Mean	Difference from control Mean difference	95% CI	s.c. histamine day 3 Mean	Difference from control Mean difference	95% CI
a Urinary histamine (ng mg^{-1} creatinine)							
Post challenge:	7.7	19.1	11.4	(-6.1 , 28.9)	22.9	15.2	(0.8, 29.5)
0–6 h:	7.7	11.2	3.5	(-9.1 , 16.1)	13.5	5.9	(-7.3 , 19.0)
6–12 h:	8.0	6.0	-1.9	(-7.2 , 3.4)	19.4	11.5	(-6.1 , 29.0)
12–18 h:	4.2	5.5	1.2	(-3.9 , 6.4)	11.6	7.4	(-4.5 , 19.2)
18–24 h:	7.7	4.2	-3.5	(-10.6 , 3.5)	8.8	1.0	(-9.3 , 11.4)
b Urinary 1-methylhistamine (ng mg^{-1} creatinine)							
Post challenge:	99.2	337.0	237.8	(60.9, 414.8)	279.2	180.0	(7.4, 352.6)
0–6 h:	99.2	225.5	126.3	(-2.1 , 254.8)	212.3	113.1	(17.3, 208.9)
6–12 h:	105.5	191.1	85.6	(-52.7 , 223.8)	226.3	120.8	(-6.0 , 247.5)
12–18 h:	133.8	143.2	9.3	(-93.5 , 112.2)	110.8	-23.0	(-111.8 , 65.8)
18–24 h:	110.8	127.6	16.8	(-58.1 , 91.6)	120.0	9.2	(-48.9 , 67.3)

Table 2 Group mean urinary histamine (Table 2a) and 1-methyl-histamine (Table 2b) excretion rates (ng min⁻¹ over 24 h periods on the 3 study days. Day 1 control, day 2 histamine bronchial provocation test (4–64 mg ml⁻¹), day 3 subcutaneous histamine injection (1 mg). All mean differences are expressed relative to the control day

	Control day 1 Mean	Inhaled histamine day 2 Mean	Difference from control Mean difference	95% CI	s.c. histamine day 3 Mean	Difference from control Mean difference	95% CI
a Urinary histamine excretion rate (ng min⁻¹)							
Post challenge:	13.8	121.9	108.1	(3.4, 212.7)	51.0	37.2	(-7.5, 81.9)
0–6 h:	13.8	11.0	-2.8	(-17.9, 12.4)	24.0	10.2	(-19.0, 39.4)
6–12 h:	13.5	13.7	0.2	(-10.8, 11.2)	17.4	3.9	(-21.2, 29.1)
12–18 h:	11.8	12.1	0.3	(-10.3, 10.9)	10.5	-1.2	(-14.7, 12.2)
18–24 h:	9.6	8.3	-1.3	(-9.6, 7.1)	6.3	-3.3	(-12.3, 5.7)
b Urinary 1-methylhistamine excretion rate (ng min⁻¹)							
Post challenge:	221.7	1862.7	1641.0	(377.6, 2904.4)	591.3	369.7	(-17.8, 757.2)
0–6 h:	221.7	195.8	-25.8	(-181.0, 129.4)	351.3	129.7	(-60.8, 320.1)
6–12 h:	233.2	306.5	73.3	(-169.8, 316.4)	370.3	137.2	(-69.4, 343.7)
12–18 h:	228.8	213.5	-15.3	(-168.9, 138.3)	222.5	-6.3	(-164.8, 152.1)
18–24 h:	234.2	243.3	9.2	(-139.1, 157.5)	198.5	-35.7	(-169.4, 98.0)

Urinary histamine/1-methylhistamine excretion rate (Table 2)

Urinary histamine excretion rate varied widely between subjects throughout the 3 study days. The mean ($\pm$ s.d.) control day 0–6 h value was 13.8 ± 14.3 ng min⁻¹ (range = 0.6–36.8 ng min⁻¹). There was a significant increase of 108 ng min⁻¹ (95% CI: 3.4, 212.7; $P = 0.04$) to 122 ng min⁻¹ in mean histamine excretion in the day 2 (after inhalation) post-provocation sample. In the 0–6 h period, the mean rate returned to a value comparable with that in the same period on day 1. Mean histamine excretion rate post-provocation after injection on day 3 was higher by 37 ng min⁻¹ (95% CI: -7.5, +81.9; $P = 0.09$) than that over the 0–6 h period on day 1.

The mean ($\pm$ s.d.) day 1, 0–6 h urinary 1-methyl-histamine excretion rate was 222 ± 144 ng min⁻¹ (range = 99–478 ng min⁻¹). There was a significant increase of 1641 ng min⁻¹ (95% CI: 377.6, 2904.4; $P = 0.02$) to 1863 ng min⁻¹ on day 2 post-provocation. In the 0–6 h interval and thereafter, excretion rates had returned to values comparable with day 1 excretion rates. There was an increase in day 3 post-injection secretion rate which was sustained over the 0–6 and 6–12 h intervals.

Discussion

The pharmacological effects of histamine observed in this study varied considerably according to the route of administration. Both routes resulted in a peak plasma concentration of histamine of 1.4–1.6 ng ml⁻¹ and this was associated with flush in all subjects. These concentrations are at variance with those reported by Kaliner *et al.* (1981, 1982) who reported that much higher concentrations are required before systemic effects appear.

However, subjects studied by Kaliner *et al.* (1981, 1982) had higher basal concentrations (0.62 ± 0.12 ng ml⁻¹) compared with those observed in the present (< 0.3 ng ml⁻¹) and in many other studies. Histamine administered subcutaneously produced a pulsatile temporal headache similar to that which occurs following an intravenous infusion (Pollock *et al.*, 1991), but had little effect on blood pressure or heart rate. In contrast, histamine given by inhalation produced no headache, minor changes in blood pressure, but did produce tachycardia. Inhalation also produced a drop in pEFR₇₀ of 45% with cough and tight chest.

There may be several reasons why symptoms differ according to the route of administration of histamine when the circulating concentrations are similar. For example, bronchial provocation tests with histamine at concentrations of 64 mg ml⁻¹ have been shown to induce cough by stimulating irritant receptors in the lungs (Lowry *et al.*, 1987), which can be blocked both by ipratropium bromide and bronchodilators. There is a further reflex component arising from irritation of afferent vagal nerve endings (Nadel & Barnes, 1984).

A high local concentration of histamine in the pulmonary circulation after histamine inhalation might have a disproportionately greater effect on the carotid artery baroreceptors than intravenous or subcutaneous injection. Also, there might be a greater direct cardiac effect mediated by stimulation of cardiac H₂ receptors thereby increasing diastolic depolarisation of the S.A. node (Douglas, 1985).

The pulsatile headache which is the most common limiting end-point for studies of the clinical effects of infused histamine (Kalinin *et al.*, 1982) occurred in our subjects after subcutaneous injection but not after inhalation. It is possible that this may reflect a difference in the rate of change in plasma histamine after stepwise inhalation relative to injected histamine. We found a

greater rate of change of histamine concentrations after subcutaneous injection (i.e. 0.3 to 1.4 ng ml⁻¹, 5 min after infusion compared with 0.3 to 1.65 ng ml⁻¹ over the 25 min of the bronchial challenge), but a much slower rate of decay. Differences in rate of decline of plasma histamine were also reflected in urine concentration.

While we have demonstrated that clinical signs and

symptoms following histamine administration differ with the route of administration, they have been shown to mimic some of the features of clinical challenges, either oral or inhaled, with foods, drugs, additives, allergens or physical stimuli. Measurements of urinary histamine and 1-methylhistamine concentration and excretion rates may be useful non-invasive markers of mediator release following allergen challenge.

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