Circulating plasma concentrations of atrial natriuretic peptide and catecholamines in response to maximal exercise in normal and asthmatic subjects

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Abstract

Background Intravenous infusion of atrial natriuretic peptide has been shown to cause bronchodilatation in patients with asthma and endogenous atrial natriuretic peptide is known to rise with exercise. Whether an aberration in release of atrial natriuretic peptide is concerned in the pathogenesis of exercise induced bronchoconstriction has not been studied.

Methods The atrial natriuretic peptide response to exercise was studied in eight men with exercise induced asthma and eight age matched non-asthmatic men. Subjects exercised to exhaustion on a treadmill, using the Bruce protocol. Atrial natriuretic peptide and catecholamines were measured at the end of each stage of exercise and oxygen consumption and heart rate were monitored throughout.

Results Both groups showed a 3-5 fold increase in plasma atrial natriuretic peptide during exercise (mean (SE): normal subjects 25 (4) pmol/l; asthmatic subjects 24 (5) pmol/l), with no difference between the two groups. There was a close correlation between plasma atrial natriuretic peptide concentrations and oxygen uptake, catecholamine release, and heart rate in both groups. The catecholamine response was similar in the asthmatic and normal subjects, both groups showing a four fold rise in plasma adrenaline and a 4-5 fold rise in plasma noradrenaline.

Conclusion A defect in the release of circulating atrial natriuretic peptide does not account for exercise induced asthma; the concentrations of the circulating peptide that were achieved may effect a small reduction in airway reactivity. Our data do not support the idea that asthmatic patients have abnormal sympathoadrenal activity.

Several studies have examined the response of atrial natriuretic peptide to exercise in normal subjects and have found plasma concentrations to increase significantly, though the mechanisms underlying this change are not clear. Atrial stretch is the predominant stimulus for atrial natriuretic peptide release and the increase in central blood volume associated with exercise is likely to have a major role. The mechanism may be affected by disease states that accentuate changes in pulmonary vascular resistance with exercise, and the increase in right heart pressures would be expected to augment atrial natriuretic peptide release yet further.

Atrial natriuretic peptide transcripts have been detected in lung tissue and the biologically active 28 amino acid form is released from isolated perfused lung, suggesting that atrial natriuretic peptide of pulmonary origin may contribute to circulating plasma concentrations or that it may have a local autocrine role in the lung. Some animal studies have shown a direct relaxant effect of atrial natriuretic peptide on airway smooth muscle and specific receptors for atrial natriuretic peptide have recently been located in bronchial and bronchiolar muscle, suggesting that atrial natriuretic peptide may act directly on the airway as well as on the pulmonary vasculature.

We have recently shown that the intravenous infusion of atrial natriuretic peptide reduces bronchomotor tone in the asthmatic and the normal human airway, and that lesser changes in plasma concentrations decrease airway reactivity in the absence of frank bronchodilatation.

These various factors suggest that circulating atrial natriuretic peptide may have a role in the airway response to exercise. Normal subjects often show a degree of bronchodilatation immediately after exercise, whereas patients with exercise induced asthma develop bronchoconstriction with a similar time course. An impaired atrial natriuretic peptide response might therefore have a detrimental effect on airway reactivity whereas an exaggerated response could be beneficial.

Strenuous exercise also causes a rise in plasma arginine-vasopressin and catecholamines, both of which have been shown to stimulate atrial natriuretic peptide secretion in vivo. Some previous studies have suggested that patients with asthma have a defective sympathoadrenal response to exercise. How this relates to the atrial natriuretic peptide response to exercise is unknown.

The current study was therefore designed to examine the dynamics of release of atrial...
natriuretic peptide in patients with exercise induced asthma and normal subjects during graded treadmill exercise continued until exhaustion, with concomitant measurement of oxygen consumption and catecholamine release.

Methods

Subjects
Eight atopic asthmatic men were enrolled, mean (SD) age 32·1 (9·7) years, with a baseline forced expiratory volume in one second (FEV,) of 3·25 (0·46) litres (80·1% (11·2%) predicted). All were taking regular inhaled beta agonists and six inhaled corticosteroids but had no other inhaled medication. Each had previously shown at least a 20% fall in FEV, after submaximal treadmill exercise but were unfamiliar with the remainder of the apparatus. The control group consisted of eight non-asthmatic men matched for age and physical activity, mean age 30·6 (5·9) years with a baseline FEV, of 4·20 (0·46) litres (100·7% (12·9%) predicted). No subject from either group was taking oral medication of any kind and all were non-smokers. The study was approved by the Glasgow West Ethical Committee and written informed consent was obtained from each subject.

Study Design
Each subject attended the laboratory once only, between 0930 and 1100 hours, having refrained from exercise for at least 24 hours. Asthmatic subjects discontinued their inhaled beta agonists for at least eight hours before the study but inhaled corticosteroids were continued as usual. On arrival an intravenous cannula (20G Viggo AB, Helsingborg, Sweden) was inserted into the right forearm and the subject was left to relax in the seated position for 20 minutes, during which time a three lead electrocardiogram (ECG) was applied. After this run in period 10 ml of venous blood were withdrawn for the estimation of basal plasma atrial natriuretic peptide and catecholamines and thereafter FEV, was recorded. The subject then mounted the stationary treadmill and was connected to the mouthpiece of the automated exercise machine. After two minutes' baseline readings exercise began according to the Bruce protocol (table 1). During the final 20 seconds of each stage of exercise 10 ml of venous blood were withdrawn for the estimation of plasma atrial natriuretic peptide and catecholamines. Each stage lasted three minutes and the subject continued until exhaustion (which was not necessarily at the end of a particular stage). Once exhaustion had been reached the subject dismounted the treadmill and resumed his seat. FEV, and plasma hormones were measured at two, five, 10 and 20 minutes after exercise. An electrocardiogram (standard lead II) was recorded throughout exercise and recovery.

Measurements

FEV, This was measured with a dry wedge spirometer (Vitalograph S, Vitalograph, Buckingham). The best of three attempts was used for analysis.

Atrial natriuretic peptide Five millilitres of venous blood were collected into potassium EDTA tubes containing aproitin 1000 KIU (TrasyloL, Bayer, Newbury), stored on ice and spun within two hours. Atrial natriuretic peptide was later measured by radioimmunoassay22 after pre-extraction of plasma on C-18 reverse phase columns (Sep-Pak, Waters, Milford, Massachusetts). Both interassay and intra-assay variation was consistently less than 10%.

Plasma catecholamines Five millilitres of venous blood were collected into lithium heparin tubes, stored on ice, and spun within two hours. Adrenaline and noradrenaline were later measured by radioenzymatic assay23 with both interassay and intra-assay variation of less than 10%.

Respiratory exercise measures Analysis of minute oxygen consumption (Vo2), carbon dioxide production, and ventilatory rate (Ve), together with heart rate and the computation of work load, was performed by an automated exercise machine (9000IV Computerised Pulmonary Exercise Laboratory, Gould Inc, Dayton, Ohio).

Data Analysis
The mean values of plasma atrial natriuretic peptide and catecholamines obtained during and after exercise were compared by analysis of variance for repeated measures. Mean values of maximal atrial natriuretic peptide plasma concentrations were compared by using the t test for paired data. Correlation coefficients were calculated by linear regression analysis. In all cases a p value below 0·05 was taken to be significant.

Table 1 Numbers of subjects reaching the different stages of the Bruce exercise protocol

<table>
<thead>
<tr>
<th>Stage</th>
<th>Speed (km/h)</th>
<th>Incline (%)</th>
<th>Number reaching stage</th>
<th>Normal subjects</th>
<th>Asthmatic subjects</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2·7</td>
<td>10</td>
<td>8</td>
<td>8</td>
<td>8</td>
</tr>
<tr>
<td>2</td>
<td>4·0</td>
<td>12</td>
<td>8</td>
<td>8</td>
<td>8</td>
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<tr>
<td>3</td>
<td>5·4</td>
<td>14</td>
<td>8</td>
<td>8</td>
<td>8</td>
</tr>
<tr>
<td>4</td>
<td>6·7</td>
<td>16</td>
<td>8</td>
<td>8</td>
<td>8</td>
</tr>
<tr>
<td>5</td>
<td>8·0</td>
<td>18</td>
<td>7</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>6</td>
<td>8·8</td>
<td>20</td>
<td>4</td>
<td>3</td>
<td>3</td>
</tr>
</tbody>
</table>

Table 2 Details of the subjects and mean (SE) peak exercise values (no significant differences)

<table>
<thead>
<tr>
<th>Variable</th>
<th>Asthmatic subjects</th>
<th>Normal subjects</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>32 (3)</td>
<td>31 (2)</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>79·1 (3·6)</td>
<td>73·6 (1·9)</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>174 (3)</td>
<td>178 (2)</td>
</tr>
<tr>
<td>Peak exercise values:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vo2 (ml/kg/min)</td>
<td>50·5 (1·9)</td>
<td>50·9 (5·8)</td>
</tr>
<tr>
<td>Ve (l/min)</td>
<td>110·4 (10·6)</td>
<td>115·4 (4·8)</td>
</tr>
<tr>
<td>Exercise time (min)</td>
<td>14·4 (0·8)</td>
<td>15·4 (0·5)</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>189 (3)</td>
<td>191 (2)</td>
</tr>
</tbody>
</table>

Vo2—oxygen consumption; Ve—ventilatory rate.
Results
Mean (SE) baseline FEV\(_1\) was 4.25 (0.25) litres (101% (5%) predicted) in normal subjects and was significantly higher than that of the asthmatic patients (3.25 (0.16) litres (80% (4%) predicted). After exercise there was a mean maximum fall of 27.1% (4.8%) FEV\(_1\) in the asthmatic subjects with a slight rise in FEV\(_1\) in the normal subjects. Details of the subjects and mean exercise values are given in table 2.

Basal plasma atrial natriuretic peptide concentrations were 7 (1) pmol/l in the normal subjects and were similar in the asthmatic subjects (7 (2) normal range 2–17 pmol/l). Atrial natriuretic peptide increased with successive stages of exercise, the maximum concentrations being recorded two minutes after completion of exercise (fig 1). Individuals peaked at slightly different times, however.

Analysis of individual maximum data showed peak concentrations of 25 (4) pmol/l in the normal subjects and 24 (5) pmol/l in the asthmatic subjects. There was a substantial variation in individual plasma atrial natriuretic peptide concentrations in response to exercise, maximal increases in both groups ranging from twofold to about 12 fold.

Throughout the exercise period plasma atrial natriuretic peptide concentration was closely correlated with oxygen uptake (r = 0.92 (normal subjects) and 0.93 (asthmatic subjects); p < 0.001). A similar correlation was found between atrial natriuretic peptide concentration and heart rate (r = 0.88 (normal subjects) and 0.87 (asthmatic subjects); p < 0.001). There was no significant correlation between atrial natriuretic peptide concentrations and change in FEV\(_1\).

Basal catecholamine concentrations were also similar in the two groups (normal subjects: adrenaline 0.5 (0.1) nmol/l, noradrenaline 2.50 (0.40) nmol/l; asthmatic subjects: adrenaline 0.5 (0.2) nmol/l, noradrenaline 2.80 (0.50) nmol/l). The catecholamine response to exercise was also similar in the two groups, showing a four fold rise in plasma adrenaline (fig 2) and a four to five fold rise in plasma noradrenaline.

Discussion
We did not find any significant difference between the response of atrial natriuretic peptide to exercise in normal and asthmatic subjects, despite exercising groups to exhaustion and inducing pronounced bronchoconstriction in the asthmatic subjects. Not only peak values but also the dynamics of release during exercise were similar. Although a significant increase from baseline concentrations was not seen in either group until stage 4, this might simply reflect the relatively short time between sampling. The plasma half life of atrial natriuretic peptide is about three minutes\(^a\) and steady state concentrations would therefore not have been achieved during individual stages of exercise.

The fact that atrial natriuretic peptide concentrations were also similar during the period of bronchoconstriction suggests that the increased pulmonary vascular resistance and thus right ventricular end diastolic pressure subsequent to airway narrowing does not alter atrial stretching sufficiently to augment atrial natriuretic peptide secretion. We have recently shown that the rise in circulating plasma atrial natriuretic peptide to the maximum concentrations seen during this study causes a modest but significant reduction in non-specific airway reactivity,\(^b\) though the maximum concentration is some 5–10 times less than that required to produce measurable bronchodilatation.\(^c\) We may postulate that without the atrial natriuretic peptide response exercise induced bronchoconstriction might have been greater.

The mean increase of about 3.5 fold in
Circulating atrial natriuretic peptide accounts well with the results of previous studies, though few protocols have required that the subject exercised to exhaustion.\textsuperscript{12,17} Recently Rubinstein \textit{et al} compared the atrial natriuretic peptide response of normal and asthmatic subjects to exercise and were also unable to find any significant difference.\textsuperscript{25} In their study 80\% of the calculated maximum heart rate had to be reached and was sustained for five minutes only on a bicycle ergometer. Atrial natriuretic peptide sampling was rather protracted and plasma catecholamines were not measured. If work loads are to be accurately matched, however, it is important that oxygen consumption is measured; other variables, such as exercise time, heart rate, and minute ventilation, give only an approximate indication of work load and are influenced by factors such as physical fitness, weight, height, and familiarity with the exercise protocol.\textsuperscript{26} In this study, because our asthmatic subjects were similar in physical fitness, height, and weight to the control group, exercise time, heart, and minute ventilation were similar between the groups (table 2). The few previous workers who have attempted to correlate oxygen consumption with atrial natriuretic peptide production during exercise\textsuperscript{12,17} have reported a close correlation. Only one group has proceeded to \(V_{O_2}\) max, however,\textsuperscript{24} and we can confirm that the correlation holds even at maximum oxygen consumption. The similarly close correlation between heart rate and plasma atrial natriuretic concentration has also been found before,\textsuperscript{1,4} but has rarely been followed to maximum heart rate. The relationship of heart rate and oxygen consumption to work load is well established,\textsuperscript{27} and the apparent correlations between atrial natriuretic peptide and these variables may simply reflect the increase in central blood volume and subsequently atrial stretch that occurs with increasing exercise. Previous studies, except for that of Richards \textit{et al},\textsuperscript{3} have assessed atrial natriuretic peptide secretion in response to exercise using a bicycle ergometer with the subjects (of necessity) in the seated or supine position. The supine position has been shown to alter the dynamics of atrial natriuretic peptide release.\textsuperscript{5,7}

Sympathoadrenal activity in patients with asthma remains the subject of considerable debate.\textsuperscript{28} It has been suggested that asthmatics have an impaired catecholamine response to exercise\textsuperscript{21} and other bronchoconstriction provocations, though this view has been challenged.\textsuperscript{28} Our results clearly showed no significant difference between normal and asthmatic subjects in either the peak plasma catecholamine concentrations or the response to increasing exercise. These findings confirm similar data reported by our group\textsuperscript{29} and by others,\textsuperscript{30} and do not support the concept of a deficit in the initial mobilisation of catecholamin in asthma.\textsuperscript{31} All the reports confirm that work load is of paramount importance in determining the sympathoadrenal response to exercise and this might explain the apparent conflict between studies. In the study by Barnes \textit{et al}\textsuperscript{30} the asthmatic subjects had a higher maximum heart rate and lower maximum ventilatory response than the control subjects, suggesting a subtle but important difference in the work loads achieved. In the other conflicting study there was even less standardisation of work loads and also a delay in the sampling of catecholamines after exercise.\textsuperscript{21}

In summary we therefore have found no significant difference between the atrial natriuretic peptide response to maximal exercise in normal and asthmatic subjects despite careful matching of work loads. We conclude that a defective atrial natriuretic peptide response is unlikely to account for exercise induced asthma, though the increased plasma concentrations may have some mild broncho-protective effect.

We are indebted to Jacqueline Doyle and Morven Campbell for their help with hormone assays. The cost of these assays was borne by the Chest, Heart, and Stroke Association (Scotland). GH was supported by a Wellcome medical graduate fellowship.