Platelet activation during exercise induced asthma: effect of prophylaxis with cromoglycate and salbutamol

CE JOHNSON, PW BELFIELD, SDavis, NJ COOKE A SPENCER, JA DAVIES
From the Department of Respiratory Medicine and University Department of Medicine, General Infirmary, Leeds

ABSTRACT Peak expiratory flow (PEF) and plasma concentrations of platelet factor 4 and β thromboglobulin were measured before and after exercise in nine asthmatic patients and 12 non-asthmatic volunteers. Exercise was preceded by administration in random order of either placebo, salbutamol 200 μg, or sodium cromoglycate 2 mg from a pressurised inhaler. In control subjects there were minimal changes in PEF and plasma concentrations of platelet factor 4 and β thromboglobulin. In the asthmatic patients the typical changes in PEF were seen on exercise; plasma concentrations of platelet factor 4 and β thromboglobulin rose significantly in parallel, the rise preceding the fall in PEF. The changes in peak flow and platelet activation induced by exercise were attenuated by prior administration of salbutamol or cromoglycate. These results indicate that exercise induced asthma is associated with a rise in platelet release products similar to that observed in antigen induced asthma.

There is considerable evidence that inflammatory mediators are the underlying agents of bronchoconstriction in asthma. Several of these substances, and particularly platelet activating factor (PAF-acether), also cause activation of blood platelets. This has been detected during antigen induced asthma by measurement of platelet factor 4, β thromboglobulin, and change in the platelet aggregate ratio. These findings have been interpreted to indicate that antigen induced asthma is accompanied by sensitisation of mast cells and release of chemical mediators. While there is general support for the idea that this pathway is concerned in antigen induced asthma, there is controversy about the role of mediators in exercise induced asthma. We have therefore assessed platelet activation during exercise in asthmatic patients known to be susceptible to exercise induced asthma by measuring platelet factor 4 and β thromboglobulin. The results obtained in these subjects were compared with findings in non-asthmatic subjects undergoing similar exercise. On separate occasions the platelet release response to exercise was measured in the asthmatic subjects after administration of sodium cromoglycate and salbutamol, drugs known to modify exercise induced asthma.

Methods
We studied nine asthmatic subjects attending the outpatient clinic (mean age 23 years) and 12 non-asthmatic members of the medical and laboratory staff (mean age 31 years). The details of the asthmatic patients are shown in the table. All subjects gave full informed consent to take part and the study was approved by the Leeds Western District research ethics committee. Both patients and control subjects discontinued all medication on the evening before each study day.

Exercise was carried out on a treadmill at room temperature (about 20°C) and all of the participants had experienced the procedure at least once before the study. The speed and elevation of the treadmill were adjusted for each individual so that six minutes of sustained submaximal exercise could be carried out. The chosen work load raised the mean pulse rate to 152 beats per min in the asthmatic subjects and 148 beats per min in the controls. Control subjects were

Address for reprint requests: Dr PW Belfield, Department of Respiratory Medicine, General Infirmary, Leeds LS1 3EX.

Accepted 24 September 1985
Platelet activation during exercise induced asthma

Peak flow meter. At the same times blood samples were collected from an antecubital vein without venous stasis. A standard procedure regularly used in the laboratory was followed to minimise activation of platelets during collection and processing of blood. Venipuncture was carried out with minimal trauma and avoidance of suction in the syringe, a 19 G Butterfly cannula being used. The first 2 ml of blood was taken into a syringe and discarded. A second syringe was used to collect the next 5 ml of blood, which was immediately and carefully decanted into an ice cooled test tube containing EDTA, theophylline, and prostaglandin E1 to inhibit platelet activation. Blood was mixed by gentle inversion of the tube and the samples were kept in melting ice for 30 minutes, before centrifugation at 2200 g and 4°C for 30 minutes. Aliquots of platelet poor plasma were removed from the middle layer of the supernatant plasma and stored at −70°C until they were assayed. Plasma concentrations of platelet factor 4 and β thromboglobulin were measured by radioimmunoassay with the kits supplied by Abbott Laboratories and Amersham International respectively. The normal ranges for our laboratory, obtained from 20 normal subjects aged 22–60 years, are 3.9 (SEM 0.3) and 31.7 (2.1) ng/ml respectively. For platelet factor 4 the interassay coefficient of variation was 5% and the intra-assay coefficient of variation 8%. For β thromboglobulin the values were 10% and 8%.

Differences between group values were assessed for statistical significance with the Wilcoxon signed rank test and the two sample test for non-parametric data.

Results

Both asthmatic and control subjects exercised to a similar extent, with mean (SEM) increases in pulse rate of 69 (4) and 70 (5) beats per minute respectively. Exercise did not cause any distress to patients or normal subjects.

Exercise did not greatly affect PEF or platelet release proteins in the control subjects (fig 1). There was a small, transient rise in the values of all three measurements after exercise but this attained significance only for PEF (p < 0.05).

Individual results for platelet factor 4 and β thromboglobulin in the asthmatic subjects before and after exercise are shown in the table. The individual rises in platelet factor 4 and β thromboglobulin were significantly correlated r = 0.83 (p < 0.01).

In the asthmatic subjects PEF measurements showed typical exercise induced bronchoconstriction (fig 1). There was no significant change immediately after exercise but 10 minutes afterwards there was a significant mean fall of 114 l min−1 (p < 0.01) below the resting value, and PEF had not returned to nor-

Fig 1 Changes in PEF and plasma concentrations of platelet factor 4 (PF4 ■■■■) and β thromboglobulin (βTG; ○○○○) during and after exercise in nine asthmatic patients and 12 normal controls (means with standard errors). The six minute exercise period is indicated by the hatched area. The significance of differences between resting and subsequent measurements (Wilcoxon test) is indicated by *(p < 0.05) and **(p < 0.01).

exercised on only one occasion, preceded by administration of two puffs from a placebo inhaler. In the asthmatic subjects exercise was carried out on three occasions at the same time of day at weekly intervals. After baseline measurements salbutamol 200 μg, sodium cromoglycate 2 mg, or placebo were administered in the form of two puffs from a pressurised inhaler. The drugs were administered according to a random allocation by one observer (CEJ). All measurements of respiratory function and sampling of blood were carried out without knowledge of which drug a particular subject had received.

After a standard period of rest for 15 minutes, laboratory measurements were made 10 minutes before the exercise period. They were repeated immediately after exercise and 10 and 25 minutes after exercise. Peak expiratory flow was measured with a Wright's
Fig 2  Effect of pretreatment with sodium cromoglycate or salbutamol on changes in peak expiratory flow (PEF) and plasma concentrations of platelet factor 4 (PF4) and β thromboglobulin (βTG) induced by a six minute period of exercise (hatched area) in nine asthmatic patients (means with standard errors). The significance of differences from resting values (Wilcoxon test) is indicated by *(p < 0.05) and **(p < 0.01).

Clinical details of asthmatic patients and the platelet factor 4 (PF4) and β thromboglobulin (βTG) responses to exercise after placebo

<table>
<thead>
<tr>
<th>Age</th>
<th>Sex</th>
<th>Atopy</th>
<th>Serum IgE* (IU/ml)</th>
<th>Mean resting (% PEF pred)</th>
<th>PF4 (ng/ml)</th>
<th>βTG (ng/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Basal</td>
<td>Peak on exercise</td>
</tr>
<tr>
<td>23</td>
<td>M</td>
<td>+</td>
<td>380</td>
<td>84</td>
<td>4</td>
<td>10</td>
</tr>
<tr>
<td>23</td>
<td>M</td>
<td>+</td>
<td>130</td>
<td>93</td>
<td>11</td>
<td>24</td>
</tr>
<tr>
<td>31</td>
<td>M</td>
<td>+</td>
<td>285</td>
<td>76</td>
<td>5</td>
<td>30</td>
</tr>
<tr>
<td>20</td>
<td>F</td>
<td>+</td>
<td>ND</td>
<td>82</td>
<td>7</td>
<td>11</td>
</tr>
<tr>
<td>24</td>
<td>M</td>
<td>+</td>
<td>95</td>
<td>60</td>
<td>6</td>
<td>27</td>
</tr>
<tr>
<td>24</td>
<td>F</td>
<td>+</td>
<td>10</td>
<td>69</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>20</td>
<td>M</td>
<td>+</td>
<td>660</td>
<td>73</td>
<td>7</td>
<td>14</td>
</tr>
<tr>
<td>20</td>
<td>F</td>
<td>+</td>
<td>300</td>
<td>85</td>
<td>12</td>
<td>12</td>
</tr>
<tr>
<td>20</td>
<td>F</td>
<td>+</td>
<td>330</td>
<td>70</td>
<td>7</td>
<td>8</td>
</tr>
</tbody>
</table>

*Normal range of IgE up to 120 IU/ml.
ND—not done; PEF—peak expiratory flow.
Platelet activation during exercise induced asthma

It is generally agreed that chemical mediators play a part in antigen induced asthma,\textsuperscript{18–21} but the evidence for their role in exercise induced asthma has been much less convincing.\textsuperscript{22–24} The present study indicates that platelet activation occurs after exercise in asthmatic patients to much the same extent as after antigen challenge.\textsuperscript{2} The findings are consistent with the view that exercise induced asthma, at least in part, is produced by chemical mediators.

We are grateful to Miss KM Milner for secretarial assistance. We thank Allen and Hanburys Ltd and Fisons Pharmaceuticals for a grant towards the costs of laboratory reagents.

References


Platelet activation during exercise induced asthma: effect of prophylaxis with cromoglycate and salbutamol.

C E Johnson, P W Belfield, S Davis, N J Cooke, A Spencer and J A Davies

Thorax 1986 41: 290-294
doi: 10.1136/thx.41.4.290

Updated information and services can be found at:
http://thorax.bmj.com/content/41/4/290

These include:

Email alerting service

Receive free email alerts when new articles cite this article. Sign up in the box at the top right corner of the online article.

Notes

To request permissions go to:
http://group.bmj.com/group/rights-licensing/permissions

To order reprints go to:
http://journals.bmj.com/cgi/reprintform

To subscribe to BMJ go to:
http://group.bmj.com/subscribe/