

Exercise-induced bronchoconstriction as an expression of bronchial hyperreactivity: a study of its mechanisms in children

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ABSTRACT The mechanism of exercise-induced bronchoconstriction (EIB) was studied by observing the protective effects of several aerosol agents in a double-blind, randomised trial. Exercise-induced bronchoconstriction was not affected by placebo, but was reduced by each agent used ($p < 0.001$). Blocking the parasympathetic system had the weakest effect, while β_2 adrenergic stimulation produced the strongest effect which was significantly different from the parasympatholytic ($p < 0.02$). The effect of the mast cell stabiliser, sodium cromoglycate (SCG) was found to be intermediate. However in some patients SCG had a stronger effect than the β_2 adrenergic agonist. A relationship was found between EIB and bronchial hyperreactivity induced by histamine ($p < 0.05$).

Bronchial hyperreactivity to nonspecific stimuli is one of the hallmarks of asthma.^{1,2} This implies that bronchoconstriction occurs in asthmatics after inhalation of given concentrations of physical or chemical agents (allergens excepted) which would have no effect on healthy subjects. The degree of hyperreactivity is commonly estimated by bronchial sensitivity to histamine, acetylcholine, or methacholine. A strong correlation has been found between bronchial sensitivity to histamine and exercise-induced bronchoconstriction,³⁻⁵ and some authors⁴ consider that EIB is an expression of bronchial hyperreactivity.

The mechanism of EIB is at present incompletely understood. The parasympathetic nervous system, activated by sensory fibres situated in the mucosa of the large airways ("irritant receptors") seems to play a role, because aerosolised parasympatholytic agents⁶ or local anaesthetics⁷ may diminish or abolish EIB. Several studies⁸ point to the important influence of rapid ventilation with cold, dry air as the initiating stimulus which may trigger irritant receptors. β_2 adrenergic agonists block EIB,⁶ which suggests that the sympathetic nervous system also plays a role. Sodium cromoglycate (SCG) often gives protection against EIB.⁹ This may indicate that

mediator release from mast cells or from related histamine-containing cells in the bronchial lumen¹⁰ is also involved. Some authors suggest that SCG may influence EIB in other ways.^{5,8,11} None of these suggestions has however been substantiated,⁵ and we have therefore assumed that the effect of SCG is brought about by mast cell stabilisation.¹²

We have measured the protective effect of various agents in a group of asthmatic children who also had EIB. A β_2 adrenergic agonist (fenoterol), a parasympatholytic agent (oxytropium bromide) as well as a mast cell stabiliser (SCG) were all tested as aerosols. In this way the contribution of the various parts of the autonomic nervous system and of mast cells in the mechanism of EIB has been studied. The bronchial sensitivity to histamine was also measured and related to the protective effect of the agents.

Methods

The group of asthmatics studied comprised nine boys and four girls (age range: 9 to 15.5 years, mean: 12.5 years). The subjects were outpatients attending the Sophia Children's Hospital in Rotterdam. The selection criteria were: initial forced expiratory volume in one second (FEV_1) $\geq 60\%$ of the mean expected value for height and exercise-induced decrease in FEV_1 $\geq 15\%$ of the initial value obtained in baseline tests on each of the three consecutive days.

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In all patients these values were reproducible—that is, their coefficient of variation (standard deviation as percentage of the mean) was less than 15%. In the two weeks before the study the patients were in a stable clinical condition and had not received SCG, beclomethasone, oral corticosteroids, or β_2 adrenergic agonists. Other drugs were withdrawn three days before the tests.

The exercise test was done on a Jaeger treadmill following the method of Eggleston and Guerrant.¹³ The slope was 10° and the running time about six minutes. The speed was increased to give a pulse rate of about 175/min. This was reached at speeds between five and 12 km/hour.

Bronchoconstriction was detected by lung function measurements using Finivess flow-volume equipment. The FEV₁ was registered by a time signal. The results were based upon the FEV₁ values. The maximal expiratory flows at 25% of the forced vital capacity were also analysed but as no significant differences were found compared with the FEV₁ values they receive no further mention.

Pulmonary function tests were performed before, immediately after and 5, 10, 15, and 20 minutes after completion of the exercise tests. At each time three flow-volume curves were recorded and the highest FEV₁ value was taken for analysis. The change in FEV₁ was expressed as a percentage of the initial value. The criterion taken for bronchoconstriction was a decrease in FEV₁ \geq 15%.

The study was carried out on four consecutive days. On the first day an exercise test was done after the administration of a placebo. On each of the three following days two exercise tests were performed, the first at 9 am without a protective agent (basal test). At 1040 am a protective agent was given and the second test was performed 20 minutes later. A two-hour interval between tests was chosen to avoid carry-over effects.¹⁴ Although uncontrolled, the temperature and humidity of the ambient air during testing were reasonably constant at approximately 22°C and 70% respectively.

On each of the test days, one of the following protective agents was given in random order: oxytropium bromide (parasympatholytic), 0.02 mg per dose aerosol (two puffs); disodium cromoglycate (mast cell stabiliser), 20 mg as aqueous solution via ultrasonic nebuliser (Monaghan 670). The use of a solution avoided cromoglycate acting by hygroscopic effect alone;¹¹ fenoterol (β_2 adrenergic agonist), 0.4 mg per dose aerosol (two puffs).

The inhalation was performed as follows: after a submaximal expiration, the patient inspired synchronously with the aerosol, the mouth being kept closed over the inhaler opening. During this inspiration the tongue was kept on the floor of the mouth.

After inspiration the breath was held for five seconds. The technique was learned using a placebo. The doses were chosen to obtain a maximal pharmacological effect.¹⁵⁻¹⁷

The histamine threshold was determined by means of the inhalation of histamine acid phosphate in increasing concentration (doses 1, 2, 4, 8, 16, 32, and 64 mg/ml).¹⁸ At the beginning of the inhalation each patient performed a submaximal vital capacity manoeuvre, after which they inhaled the aerosol at tidal volume for 30 seconds. The time between the inhalations was three to five minutes. The histamine concentration which caused a decrease in FEV₁ \geq 15% of the initial value was called the histamine threshold. With this technique the histamine threshold in healthy children is \geq 32 mg/ml.

An analysis of variance was done in order to detect any influences of day or agent sequence on the initial FEV₁ (every day at 9 am and during the day) and the decrease of the FEV₁ induced by exercise without protection (basal tests). The effects of the protective agents and comparisons between them were analysed using paired Student *t* tests. The relationship between the decrease in FEV₁ without agents and the degree of protection by each agent as well as the histamine threshold were analysed by linear regression equations from which *p* values were derived. The patient groups were divided according to the greatest effect by paired comparison between the three agents. These patient groups were compared according to initial FEV₁, decrease in FEV₁ induced by exercise without protection and the histamine threshold. The differences were analysed by Wilcoxon tests. Five per cent was taken as the level of significance.

All patients and their parents were fully informed about the aims and nature of the study and gave their consent.

Results

In the basal tests there were no correlations between day and agent sequences on the one hand and the initial FEV₁ and the decrease in FEV₁ in the basal tests on the other (analysis of variance). This makes the influence of variables other than the protective agents on the results improbable. The initial FEV₁ values at 9 and 11 am did not change significantly except after the administration of the β_2 adrenergic agonist when they were just significantly higher (*p* < 0.05).

The table shows the mean change in FEV₁ after exercise both with and without protection for the group as a whole. The decrease in FEV₁ without protection was 32.4 \pm 3.0% and the results after placebo did not differ significantly. After admini-

Table Mean changes in FEV₁ induced by exercise with and without placebo and each of the protecting agents

Agents used	Decrease in FEV ₁ induced by exercise		Degree of protection†		Difference between agents p value
	Mean ± SEM	p value	Mean ± SEM	p value	
None	-32.4 ± 3.0				
Placebo	-27.5 ± 3.2	NS			
Parasympatholytic	-12.5 ± 4.7	<0.001	16.6 ± 4.1	<0.01	$\left. \begin{array}{l} \cdot \\ \cdot \end{array} \right\} \text{NS}$ $\left. \begin{array}{l} \cdot \\ \cdot \end{array} \right\} \text{NS}$ $\left. \begin{array}{l} \cdot \\ \cdot \end{array} \right\} <0.02$
Mast cell stabiliser	-6.7 ± 2.1	<0.001	25.8 ± 4.1	<0.001	
β ₂ adrenergic agonist	+1.5 ± 5.2	<0.001	32.5 ± 5.4	<0.001	

Mean ± SEM are expressed as percentage of the initial values.
 †Difference of change in FEV₁ with and without agents.

stration of the protective agents the fall in FEV₁ was reduced. The changes in FEV₁ after each protective agent were significantly different from those without (p < 0.001). The degree of protection, defined as the difference between the decrease in FEV₁ with and without agents, was significant for each agent. When the effects of the protective agents were compared with one another significant differences were observed only between the parasympatholytic and the β₂ adrenergic agonist (p < 0.02) (fig 1).

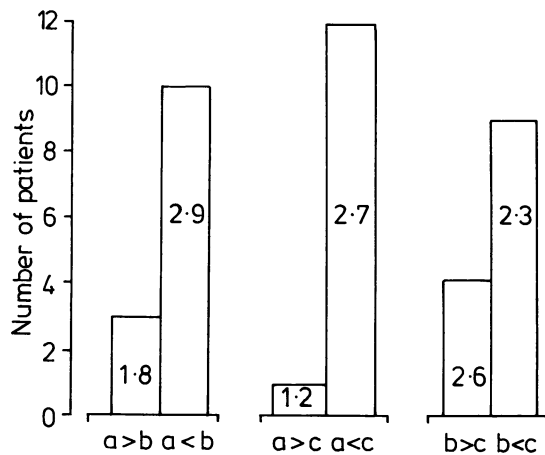


Fig 1 Number of patients according to best degree of protection in EIB by paired comparison of the agents. a = parasympatholytic; b = mast cell stabiliser; c = β₂ adrenergic agonist. The difference in effect is expressed for each patient as the quotient of the degree of protection by the agents compared. The medians of the quotients are shown in the columns.

A check was made to see if individual patients reacted differently from the group as a whole. For each patient the protective agent which gave the best degree of protection was determined by paired comparison between the agents (fig 2). The parasympatholytic gave better protection in three patients by comparison with the mast cell stabiliser and in one patient when compared with the β₂

adrenergic agonist. The mast cell stabiliser gave better protection than the β₂ agonist in four patients.

For each patient differences in protective effect were expressed in quantitative terms as the quotient of the degree of protection of the agents compared. Nearly all quotients are markedly greater than 1, indicating that the differences in degree of protection are relevant. In fig 2 decrease in FEV₁ induced by exercise without protection is compared with degree of protection afforded by the parasympatholytic and the mast cell stabiliser. The cumulative degree of protection of both agents equates with exercise induced fall in FEV₁ (difference less than 10%) in nine patients and exceeds it in the remaining four patients. Figure 2 also shows great individual differences between the cumulative degree of protection given by the parasympatholytic and mast cell stabiliser and the degree of protection given by the β₂ adrenergic agonist.

Figure 3 shows that the relationship between the decrease in FEV₁ induced by exercise without protection and the histamine thresholds is significant (p < 0.05). No relationship could be detected between the decrease in FEV₁ induced by exercise and the initial FEV₁ (r = 0.40, not significant).

In fig 4 the patients are grouped according to the agent which gave the best protection by paired comparison. No significant differences between the groups existed in the initial FEV₁, decrease in FEV₁ after unprotected exercise or histamine thresholds (Wilcoxon tests).

Discussion

In this study the protective effects of a parasympatholytic, a mast cell stabiliser (SCG) and a β₂ adrenergic agonist on EIB were found to be significant. It is assumed that maximal bronchodilatation was achieved by the doses used and that the same was true for the protective effect on EIB although this remains unproven. Although the time between administration of the parasympatholytic and the exercise test was a little short and bronchodilatation might have occurred during the recovery

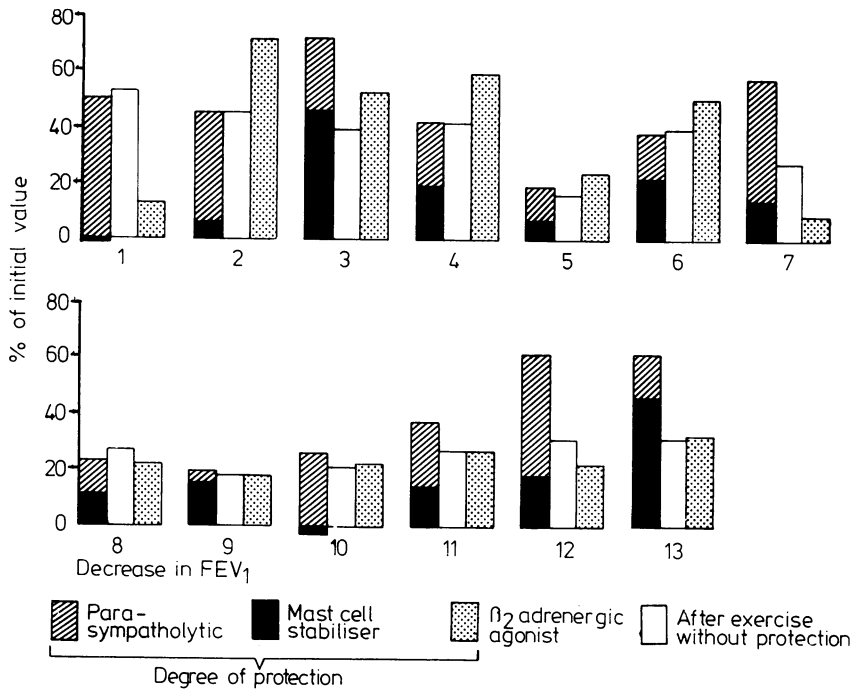


Fig 2 Degree of protection in EIB in the 13 patients given by the parasympatholytic and the mast cell stabiliser, illustrated as cumulative effects, compared with that of the β_2 adrenergic agonist and the exercise-induced decrease in FEV₁ without protection. Both degree of protection and decrease in FEV₁ are expressed in percentages of the initial FEV₁.

phase, we do not think this could have influenced the results significantly. The results were not influenced by variables other than the protective agents used and they are in accordance with those of other authors.^{6 19 20}

Individual patients differed in their responses to the various agents used. From the finding in some patients that the degree of protection given by the parasympatholytic exceeded that given by the mast cell stabiliser, it may be concluded that in these subjects activation of the vagus is more important than activation of the mast cell system. In contrast to the results of the patient group in general, some patients attained a greater degree of protection from the mast cell stabiliser than from the β_2 adrenergic agonist. In these patients it can be assumed that mediator release from mast cells is relatively important in the mechanism of EIB. It seems that blocking of histamine release from their mast cells by triggering the β_2 adrenergic receptors is only of limited value in contrast to the effect of SCG. Also, β_2 adrenergic relaxation of their bronchial muscles is of relatively little importance in the blocking of the EIB response. Although this might indicate hypo-responsiveness of their β_2 adrenergic receptors, this is unlikely because the bronchodilatation induced by

the β_2 adrenergic agonist under resting conditions in these patients was not less than in the others.

Our finding of a relationship between the degree of protection given by the parasympatholytic together with the mast cell stabiliser and the degree of bronchoconstriction without protection can be explained in two ways. Firstly, a combined mechanism involving both the parasympathetic nervous system and mast cells may operate in EIB. It was not possible to predict which was the dominant system

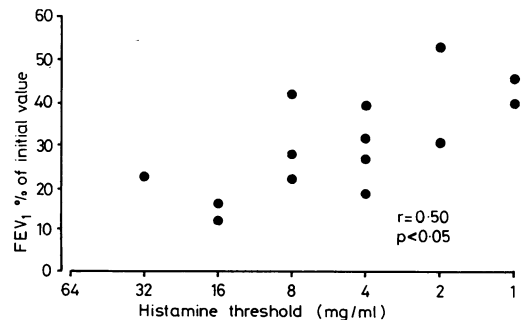


Fig 3 Relationship between the histamine threshold and the exercise-induced decrease in FEV₁.

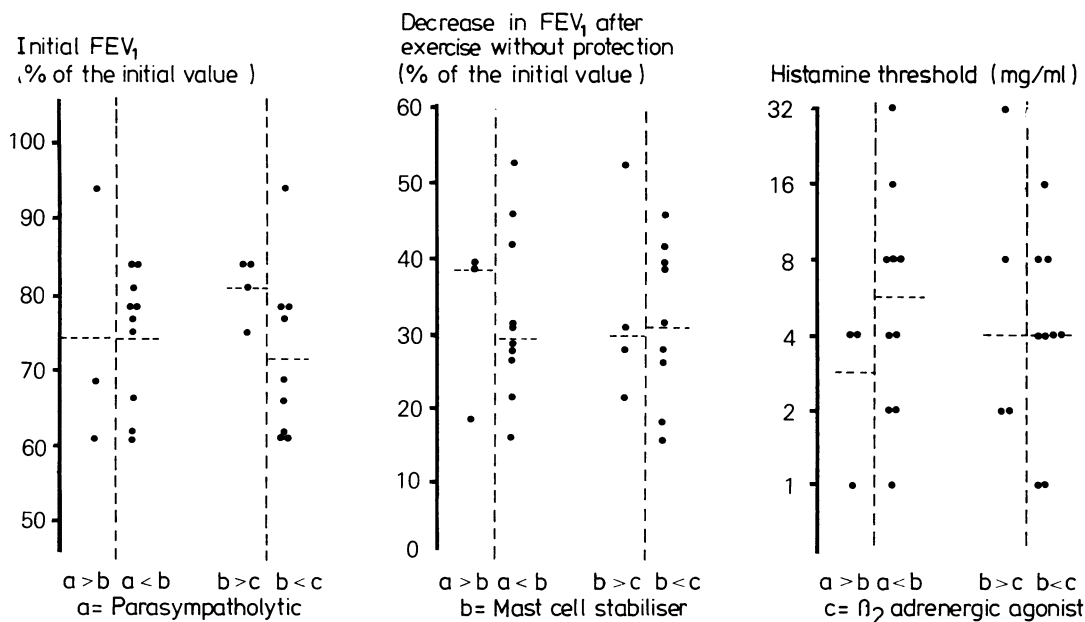


Fig 4 Initial FEV₁, decrease in FEV₁ after exercise without protection, and histamine threshold in patient groups divided according to the best protection in EIB by paired comparison of the agents. a = parasympatholytic, b = mast cell stabiliser, c = β₂ adrenergic agonist, ----- = median. Differences between groups are not statistically significant.

from the initial lung function values, the bronchodilatation induced by the agents without exercise or the bronchial sensitivity to histamine. McFadden *et al*²¹ were also able to distinguish subgroups in their population with EIB and this may well be relevant to the choice of therapeutic agent. Secondly, a complete stabilisation of the mast cell, resulting in blocking of mediator release, may only occur if in addition to cromoglycate sensitive receptors the parasympathetic receptors of the mast cell are blocked as well. If it turns out that cromoglycate has effects outside mast cells then the interpretation of blocking studies such as ours will have to be reviewed. The blocking effect of cromoglycate might then be explained in part by an action on bronchial smooth muscle or irritant receptors. The important place of the mast cells in EIB is however supported by the observations that EIB is followed by a period of latency¹⁴ and that several authors^{22 23} have found a rise in arterial histamine concentration after exercise.

The degree of bronchoconstriction induced by exercise has a strong relationship with the bronchial sensitivity to histamine as shown in this and other studies.^{4 5} This suggests that the mechanisms induced by both stimuli may be related. There is no difference in bronchial sensitivity to histamine in patients with EIB in whom the autonomic nervous

system dominates compared with those in whom mast cells are of major importance. Thus one might suspect that both these systems play a role in the mechanism leading to bronchoconstriction after histamine inhalation. Although the reaction to histamine could be confined to the direct stimulation of the histamine receptors of the bronchial muscle cells,²⁴ this is probably not the case. Jackson *et al*²⁵ showed in rats that the autonomic nervous system was stimulated by the histamine aerosol, since the bronchial response decreased after dissection of the vagus nerve. The study of Dixon *et al*²⁶ indicates that this may be caused by stimulation of irritant receptors. It seems probable that histamine-induced vagal stimulation also triggers mast cells, because the mast cell stabiliser SCG diminishes the bronchial response to histamine in some patients.^{27 28}

The mechanisms by which histamine and exercise induce bronchoconstriction are similar and this justifies the view that EIB can be regarded as an expression of bronchial hyperreactivity.

We would like to thank Jeannette Boogaard, Ineke Strik, Edith Bonzet, and Joke Mazee for their technical assistance. This study was supported by the Dutch Asthma Fund (project no 279) and Boehringer Ingelheim Ltd, Haarlem who also supplied the drugs.

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