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Clinical aspects of allergic disease

Effect of multiple doses of nedocromil sodium given after allergen inhalation in asthma

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Aim: *Twelve subjects with asthma took part in a placebo-controlled crossover study designed to investigate whether nedocromil sodium given after the occurrence of the early-phase asthmatic reaction to allergen has an effect on the late-phase response and the associated increase in airway responsiveness.*
Methods: *The treatments were administered four times at 4-hour intervals at a dose of 4 mg, with the first dose given 1 hour after the last allergen challenge. Changes in airway caliber were monitored for 15 hours after allergen exposure by measuring forced expiratory volume in 1 second hourly. Airway responsiveness to methacholine was determined 24 hours before and 24 hours after allergen challenge.*
Results: *Nedocromil sodium failed to reduce significantly the maximum late fall in forced expiratory volume in 1 second as compared with placebo but delayed its occurrence by 1.5 hours ($p = 0.05$). Nonspecific airway responsiveness to methacholine was similarly increased after allergen challenge when patients received nedocromil sodium and placebo. No unusual events were reported during the study period by any patient. These results indicate that nedocromil sodium is not able to interrupt the ongoing cascade of inflammatory events leading to the late-phase reaction and the associated increase in airway responsiveness.*
Conclusion: *In allergic asthma, nedocromil can be used only as a preventive treatment.*
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Key words: *Asthma, drug therapy, bronchial provocation test, bronchoconstriction, bronchial hyperreactivity, late-phase response*

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Inhalation of allergen causes a biphasic bronchocospastic response in many subjects with asthma.[1-3] This is constituted by a transient early-phase response (EAR), which can be reversed either spontaneously or by administration of bronchodilators, followed by a sustained late-phase response (LAR), which is long-lasting and can be reversed only in part by administration of bronchodilators. The available data suggest that

Abbreviations used
AU: Arbitrary units
EAR: Early asthmatic response
FEV_1: Forced expiratory volume in 1 second
LAR: Late asthmatic response
PD_{20}: Provocative dose causing a 20% fall in FEV_1

an inflammatory response characterized by influx of eosinophils into the airways[4, 5] is responsible for the LAR and the associated increase in airway responsiveness.[6] However, it seems that the primary phenomenon starting the cascade of these inflammatory events is the mast cell degranulation that occurs during EAR.[7]

Cromolyn sodium[8] and nedocromil sodium[9] have been shown to be capable of inhibiting both EAR and LAR when they are administered before the exposure to allergen. This protective activity is believed to be the result of their mast cell–stabilizing action.[10] When administered after EAR, cromolyn sodium was able to affect LAR and the associated increase in airway responsiveness in children[11] but not in adults,[12] whereas a single dose of nedocromil sodium tended to delay the onset of LAR also in adults.[9] These data may suggest that nedocromil sodium also has an effect on the inflammatory events that follow EAR and culminate in LAR.

The objective of this study was to investigate whether multiple doses of nedocromil sodium given after EAR may inhibit the development of LAR and the associated increase of airway responsiveness in adult patients with asthma.

METHODS

Subjects

Twelve subjects with asthma who were sensitized to house dust mite and had a biphasic response to inhalation of this allergen entered the study. Their characteristics are shown in Table I. The diagnosis of asthma was based on the criteria of the American Thoracic Society,[13] and the allergic sensitization was proved by skin prick test or RAST.

To enter the study subjects were required to be in stable condition and to have forced expiratory volume in 1 second (FEV_1) greater than 70% of predicted value[14] without circadian variations exceeding 10%. Subjects were not included in the study if they had allergic sensitization to seasonal allergens, if they had had respiratory infections in the previous 4 weeks, or if they had overt disease other than asthma. Five subjects were being treated with short-acting inhaled bronchodilators, and one was receiving cromolyn sodium. These drugs were withdrawn 6 and 48 hours, respectively, before each study day. None of the subjects were receiving theophylline, long-acting bronchodilators, antihistamines, or steroids.

TABLE I. Patient characteristics

Treatment sequence group	P/N	N/P
Sex (M:F)	5:1	2:4
Age (yr)	23 ± 11	30 ± 13
Height (cm)	168 ± 9	167 ± 1
Age at onset of asthma (yr)	8 ± 10	13 ± 9
Present Therapy		
Ipratropium + Fenoterol	1	0
Salbutamol	2	2
Sodium Cromoglycate	1	0

Data are means ± SD or numbers of subjects.
P, Placebo; *N*, nedocromil sodium.

All subjects gave their informed consent to the trial, which was conducted in accordance with the Declaration of Helsinki of 1975 and approved by the ethical committee of our department.

Bronchial challenges

Forced expiratory maneuvers were recorded by a turbine spirometer (Micro Spirometer, Micro Medical Limited, Rochester, U.K.), and the greatest value of FEV_1 from three technically acceptable maneuvers performed 1 minute apart was retained for analysis. On each day, baseline FEV_1 was obtained by averaging measurements taken 10 and 20 minutes before challenge. Methacholine or allergen aerosols were delivered by an ampule-dosimeter device (Mefar, Brescia, Italy) according to a protocol previously described.[9] Control measurements of FEV_1 were determined after inhalation of saline solution.

For methacholine challenge 1, 10, and 50 mg/ml isotonic solutions were prepared. Methacholine challenge was started at a dose of 0.02 mg (4 inhalations of a 1 mg/ml solution) with twofold increments obtained by increasing the number of breaths and concentrations until the FEV_1 measured 1 minute after inhalation was less than or equal to 80% of control. The maximum dose at which the challenge was stopped if the subject did not respond was 5 mg. Allergen bronchial challenge was performed by using scalar solutions of *Dermatophagoides pteronyssinus* (Pharmacia, Uppsala, Sweden) predosed in arbitrary units (AU) by means of RAST inhibition technique against an in-house sera pool containing a high titer of specific IgE. Fifteen minutes were allowed between allergen inhalations and FEV_1 measurements. The allergen bronchial challenge was started from a dose of 4 AU, inhaled with 50 quiet

TABLE II. Individual patient data during selection phase

Patient No.	Treatment sequence group	FEV_1 Baseline 1	FEV_1 Maximum early fall (%)	FEV_1 Maximum late fall (%)	PD_{20} - methacholine: time relative to allergen challenge Pre (μg)	Post (μg)	Ratio Post/Pre
2	N/P	3.02	41	58	48.5	14	0.28
3	N/P	2.62	55	46	149	37	0.25
4	N/P	2.91	24	31	211	74	0.35
12	N/P	3.86	38	21	73	40	0.55
13	N/P	4.26	28	54	114	36	0.32
14	N/P	2.28	28	37	47	<20	0.43
1	P/N	2.64	31	42	130	19.5	0.15
5	P/N	2.96	38	21	203	76	0.37
6	P/N	4.40	29	19	330	151	0.46
9	P/N	4.07	25	46	70	28	0.40
10	P/N	3.62	34	36	159	33	0.21
17	P/N	3.02	27	47	42	19	0.45
Mean		3.31	33	38	107	36	0.35
SD		0.71	8	12	(GM)	(GM)	0.11

N, Nedocromil sodium; *P*, placebo; *GM*, geometric mean.

breaths of a 16 AU/ml concentration, with twofold increments until the FEV_1 was less than or equal to 80% of control. The maximum dose of allergen given to subjects who did not respond was 500 AU.

Protocol

Initially, subjects entered a selection phase during which they underwent, on 3 consecutive days, a methacholine challenge (first day), an allergen challenge (second day), and a methacholine challenge (third day). All challenges were performed at the same time of the morning (i.e., 9:00). The provocative doses of methacholine and allergen causing a 20% decrease of FEV_1 (PD_{20}) were determined. Only subjects with a late-phase response (i.e., an FEV_1 fall >15% of control 3 to 12 hours after challenge) and a methacholine PD_{20} after allergen challenge less than or equal to half of that before allergen challenge proceeded to the treatment phase.

During the treatment phase, the above procedure was repeated twice in a double-blind fashion, with an interval of at least 1 week. To proceed with this phase, it was required that methacholine PD_{20} measured before allergen day was not significantly different from that measured on the first day of selection phase, that is, less than twice and more than half of the selection phase methacholine PD_{20}. In this phase the allergen PD_{20} calculated during the selection phase was to be administered. If this dose caused a fall in FEV_1 of less than 20%, the dose was repeated once. On the second day, on each occasion subjects received four doses (at four hourly intervals) of either nedocromil sodium (Tilade) (4 mg) or placebo, with the first dose given 1 hour after allergen challenge. The order in which treatments were received was randomized so that half of the subjects received nedocromil sodium first and half received placebo first. After allergen challenge and treatment administration, FEV_1 was monitored hourly until midnight.

Statistical analysis

FEV_1 was analyzed with the use of the percentage change from baseline. Analysis was performed on the maximum change in FEV_1 during LAR, the time to maximum change, the changes in FEV_1 at each time point after allergen challenge during dosing, and in the change in log-transformed methacholine PD_{20}. This was carried out by analysis of variance with subjects, treatments, and time as factors. The reproducibility of log-transformed methacholine PD_{20} between the selection and the placebo phases was assessed by calculating the mean difference and 95% confidence limits.[15] The mean geometric ratio between methacholine PD_{20} of selection and placebo phases was 0.98, and 95% confidence limits were 1.77 and 0.54. Two-tailed tests at 5% significance level were used throughout. Data are expressed as means ± SD except for PD_{20}, which is expressed as geometric mean.

RESULTS

Selection phase

Twelve of 18 subjects who entered the selection phase satisfied the criteria to be admitted to the treatment phase. In these subjects the maximum percentage fall in FEV_1 during LAR ranged from

TABLE III. Individual results of allergen challenge during treatment phase

Patient No.	FEV_1 maximum fall		Time to maximum FEV_1 fall (hr)	PD_{20} - methacholine: time relative to allergen challenge		
	Early (%)	Late (%)		Pre (μg)	Post (μg)	Ratio Post/Pre
Placebo						
2	35	48	12	51	19.5	0.38
3	37	37	12	148	17	0.11
4	21	18	9	279	145	0.52
12	20	24	6	66	22	0.33
13	33	58	13	142	36	0.25
14	30	49	11	51	<20	0.39
1	24	28	7	170	209	1.23
5	40	37	7	104	37	0.36
6	26	25	11	503	106	0.21
9	20	40	13	46	<20	0.43
10	27	14	12	148	71	0.39
17	17	56	15	53	<20	0.38
Mean	28	37	10.7*	109	40	0.41
SD	7	4	0.8	(GM)	(GM)	0.28
Nedocromil						
2	24	23	15	74	59	0.79
3	35	38	12	135	18	0.14
4	20	26	13	289	39	0.13
12	36	21	8	128	57	0.45
13	36	58	15	77	32	0.42
14	57	53	9	50	20	0.40
1	20	21	9	156	63	0.40
5	35	19	13	110	57	0.51
6	25	19	10	547	132	0.24
9	20	25	13	66	<20	0.30
10	21	24	14	118	71	0.60
17	25	60	15	73	<20	0.27
Mean	29	33	12.2*	118	40	0.39
SD	11	5	0.7	(GM)	(GM)	0.19

GM, Geometric mean.
*$p = 0.05$.

19% to 58% of control measurement. The mean ratio of preallergen to postallergen methacholine PD_{20} was 0.35 ± 0.11 (range, 0.1 to 0.5). Individual data are given in Table II.

Treatment phase

The results of allergen challenges are summarized in Table III. The mean maximum change in FEV_1 during LAR was similar with nedocromil sodium (33 ± 5) and placebo (37 ± 4). The mean time to maximum fall in FEV_1 during LAR was longer by 1.5 hours with nedocromil sodium compared with placebo ($p = 0.05$), suggesting a delay in the development of LAR with nedocromil sodium.

The time courses of changes in FEV_1 after allergen challenge with placebo and nedocromil sodium are shown in Fig. 1. The differences between the two treatments were mostly in favor of nedocromil sodium but never reached statistical significance.

The methacholine PD_{20} (geometric mean) before antigen challenge was 0.109 mg for placebo and 0.118 mg for nedocromil sodium. The mean ratio of preallergen to postallergen methacholine PD_{20} was 0.41 with placebo and 0.39 with nedocromil sodium. This difference was not statistically significant.

In only one subject (no. 9) were the doses of allergen needed to attain an early fall in FEV_1

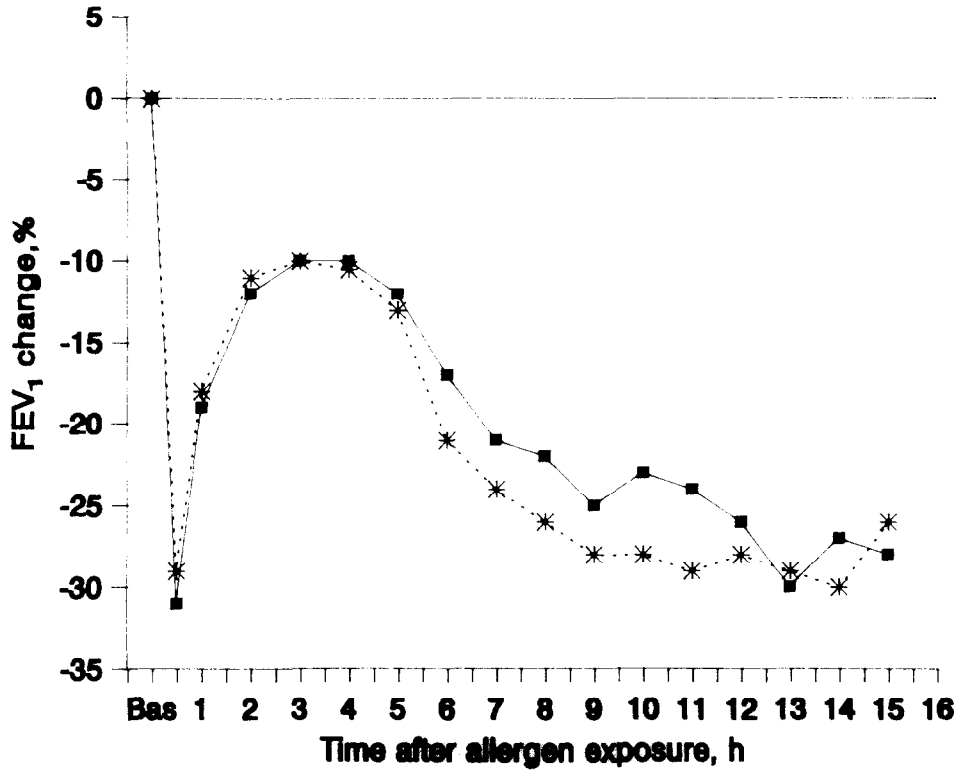

FIG. 1. Changes in FEV_1 after allergen exposure. *Asterisks* indicate placebo; *squares,* nedocromil sodium. Note that nedocromil sodium delayed the onset of late-phase asthmatic response by 1.5 hours ($p = 0.05$) even if it was unable to reduce the mean maximum fall in FEV_1 as compared with placebo. *h,* Hours.

(20%) different (16 and 32 AU) in the two treatment periods. In subjects 9, 14, and 17 on day 3 of both treatment periods, methacholine PD_{20} was not measurable because baseline FEV_1 values were too low and dropped further after administration of saline solution. This was also the case for subject 14 during the selection phase. In these cases methacholine PD_{20} was considered to be less than 0.02 mg, and this value was used for statistical analysis.

DISCUSSION

The results of this study show that nedocromil sodium given after the occurrence of early-phase response to allergen is not able to prevent the development of LAR but only alters its time course.

LAR, which often follows the experimental exposure to the sensitizing allergen, has been shown to be associated with airway inflammation.[16-18] The occurrence of LAR is also associated with an increase of bronchial responsiveness similar to that observed after the natural exposure to allergen.[19] Therefore LAR seems to represent the experimental model closest to natural asthma and appears to be useful for assessment of the antiinflammatory properties of antiasthmatic drugs.

Cromones (sodium cromoglycate and nedocromil sodium) are antiinflammatory drugs, and their best known mechanism of action is mast cell–membrane stabilization in vitro. Accordingly, studies in vivo showed that both cromolyn and nedocromil sodium are able to prevent EAR to inhaled allergen, which is known to be mainly determined by mediators released from mast cells. In addition, these drugs, when given before allergen inhalation, also inhibited the development of LAR. It is believed that this is also a result of the inhibition of the early release of mediators from mast cells, which is likely the primary mechanism starting the cascade of inflammatory events that culminate in LAR. However, the early mediator release from mast cells seems to be a necessary but not sufficient event to cause LAR because other inflammatory cells, namely eosinophils and lymphocytes,[20] must be involved.

As compared with cromolyn sodium, the more

recently developed drug nedocromil sodium has been shown in vitro to have more potent antiallergic and antiinflammatory properties[21, 22] including inhibition of eosinophil killing activity.[23] This suggests that nedocromil sodium might inhibit LAR by blocking not only the release of mast cell mediators but also eosinophil activation. An increase in the number of eosinophils in the airway was observed 4 hours after experimental inhalation of allergen, when LAR had yet to occur,[6] and this was associated with an increase in airway responsiveness to methacholine. Therefore if nedocromil sodium also acts on eosinophils in vivo, then it should also be able to inhibit the development of LAR and the associated increase in airway responsiveness when given after EAR. A previous study[9] showed that a single dose of nedocromil sodium given after EAR was not able to inhibit the development of LAR but tended to delay it, even though not significantly. From that study, however, no definite conclusion could be drawn because the duration of action of a single dose might not have been long enough to cover the whole period through which LAR usually develops.

In the present study, however, multiple doses of nedocromil sodium given after EAR failed to block the development of LAR but only tended to delay it. Moreover, nedocromil sodium failed to prevent the allergen-induced increase of bronchial responsiveness that is associated with LAR. Thus we could not confirm in adults the result of the pediatric study,[11] which showed a significant preventive effect of cromolyn given after EAR on the allergen-induced increase of bronchial responsiveness: a possible explanation for this discrepancy is the different times at which methacholine challenges were performed. The mechanism through which nedocromil sodium may delay the development of LAR is not clear. It has been shown that a second peak of mast cell mediator release occurs in blood during development of LAR.[24] It is therefore possible that the delay of LAR afforded by nedocromil sodium is related to inhibition of late mast cell degranulation. The inability of nedocromil sodium given after EAR to prevent LAR and the associated increase in airway responsiveness suggests that this drug is not active in vivo on eosinophils and other cells involved in the pathogenesis of LAR. From preliminary unpublished observations in our laboratory, it appears that inhaled steroids (beclomethasone dipropionate) 500 μg given twice at four hourly intervals, starting 1 hour after allergen inhalation, is able to fully or partially prevent the development of LAR and the associated increase in airway responsiveness. These data suggest that the antiinflammatory activity of nedocromil is different from that of inhaled steroids. This aspect deserves further investigation.

We conclude that nedocromil sodium given at a time when the inflammatory cascade is ongoing cannot completely inhibit the development of LAR but can only affect its time course. Therefore the present data confirm results of clinical studies indicating that nedocromil sodium should be used in asthma as a long-term preventive treatment only.

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A device for overcoming discoordination with metered-dose inhalers

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Background: *Despite widespread acceptance of metered-dose inhalers (MDIs) in the treatment of asthma, many patients fail to operate these devices correctly. Inability to properly coordinate activation with onset of inhalation is regarded as the major factor in suboptimal MDI therapy.*
Methods: *We evaluated Autohaler Inhalation Device (3M Pharmaceuticals, St. Paul, Minn.), a breath-activated MDI that is typically activated at a triggering flow rate of approximately 0.5 L/sec. We compared bronchodilator effect of pirbuterol acetate (Maxair), inhaled from Autohaler and a standard MDI, under conditions that ensured optimal technique in 20 patients with asthma. Spirometric variables (forced expiratory volume in 1 second [FEV_1], forced expiratory flow between 25% and 75% of vital capacity [FEF_{25-75}], forced vital capacity [FVC]) were measured before and at 15, 30, 60, and 90 minutes after two inhalations of full inspiratory reserve volume for each device.*
Results: *Both devices produced significant and similar bronchodilation. Mean FEV_1 increased 32% above baseline 60 minutes after use of Autohaler and 31% after use of a standard MDI. Similar changes were noted in FEF_{25-75} and FVC for the two devices. Differences between devices for all spirometric variables were not statistically significant.*
Conclusion: *Autohaler provides a promising alternative to the standard MDI by overcoming breath-hand discoordination. (J ALLERGY CLIN IMMUNOL 1993;92:783-9.)*

Key words: *Metered-dose inhaler, discoordination, Autohaler, breath-activated inhaler, spacers, pirbuterol acetate*

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