

Inhibitory effect of indomethacin on tachyphylaxis in response to acetaldehyde-induced bronchoconstriction in patients with asthma

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Background: Acetaldehyde, a main factor in alcohol-induced asthma, causes bronchoconstriction indirectly through histamine release; and tachyphylaxis in response to repeated inhalation of acetaldehyde is observed in patients with asthma. **Objective:** The study was designed to clarify the mechanism of tachyphylaxis in response to acetaldehyde-induced bronchoconstriction.

Methods: We investigated the bronchial response to inhaled acetaldehyde in 10 patients with asthma who were treated with indomethacin in a double-blind, randomized, placebo-controlled, crossover fashion.

Results: The mean acetaldehyde concentration causing a 20% fall in FEV₁ with placebo increased significantly from 13.0 mg/ml (geometric SEM = 0.115) to 31.1 mg/ml (geometric SEM = 0.069) over a period of 1 hour ($p < 0.01$), whereas there was a slight but not significant tachyphylaxis during indomethacin treatment. The tachyphylactic effect, expressed as logarithmic value of the second PC₂₀ minus logarithmic value of the first PC₂₀, was significantly ($p < 0.05$) reduced from 0.380 (0.066) with placebo treatment to 0.148 (0.094) with indomethacin treatment.

Conclusion: These results suggest an important role of cyclooxygenase pathway products in decreased response to repeated inhalation of acetaldehyde in patients with asthma. (*J Allergy Clin Immunol* 1997;99:620-3.)

Key words: Acetaldehyde, asthma, histamine, indomethacin, prostaglandin

Bronchial hyperresponsiveness, which is usually measured by histamine, acetylcholine, or methacholine is a main feature of bronchial asthma. Challenges with histamine and acetylcholine cause airway narrowing, mainly through direct action on airway smooth-muscle receptors; whereas exercise-, alcohol-, hyperventilation-, and cold air-induced bronchoconstriction are caused by secondary substances such as endogenously released histamine.¹⁻⁴ Although Manning et al.⁵ demonstrated, in patients with asthma, that tachyphylaxis occurs on repeated challenges with inhaled histamine but not with

Abbreviations used

GSEM:	Geometric standard error of the mean
PC ₂₀ -AcCHO:	Acetaldehyde concentration causing a 20% fall in FEV ₁

inhaled acetylcholine, Ruffin et al.⁶ reported no change in bronchial responsiveness to inhaled histamine with repeated testing. Currently, although the occurrence of tachyphylaxis in response to exogenously administered histamine has not been clearly established, we have demonstrated that tachyphylaxis in response to histamine is observed only when histamine is released endogenously after inhalation of acetaldehyde.⁷

Acetaldehyde, a rapidly generated metabolite of ethanol, is thought to be a main factor in alcohol-induced asthma. Findings indicating that alcohol-induced asthma, which occurs in response to acetaldehyde-produced histamine, is a condition that occurs only as the result of a genetic alteration in the metabolism of alcohol are that: (1) blood acetaldehyde and histamine concentrations increase during alcohol-induced bronchoconstriction⁸; (2) inhaled acetaldehyde provokes bronchoconstriction indirectly through histamine release in guinea pigs⁹ and patients with asthma¹⁰; (3) both bronchoconstriction induced by inhalation of acetaldehyde¹⁰ and acetaldehyde-induced bronchial hyperresponsiveness¹¹ occur not only in patients with so-called alcohol-induced asthma but also in virtually all patients with asthma; (4) the activity of aldehyde dehydrogenase, which oxidizes acetaldehyde to acetate, is a major factor in determining exacerbations of asthma after alcohol intake¹²; (5) among patients with asthma in Western societies, who usually do not experience bronchoconstriction in response to alcohol, severe asthma attacks follow alcohol intake in those with chronic alcoholism who are taking the aldehyde dehydrogenase inhibitor disulfiram as a component of alcohol abstinence therapy¹³; (6) terfenadine, a selective histamine H₁-antagonist, completely inhibits alcohol-induced bronchoconstriction.²

On the other hand, we have reported that thromboxane A₂ is involved in acetaldehyde-induced bronchoconstriction in patients with asthma.¹⁴ These observations prompted us to consider that local production of inhib-

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itory prostaglandins might play a part in tachyphylaxis in response to repeated challenges with acetaldehyde. Furthermore, no study has, to our knowledge, examined the role of prostaglandins in tachyphylaxis in response to bronchoconstriction mediated by endogenous histamine. We therefore examined the effect of indomethacin, a cyclooxygenase inhibitor, on tachyphylaxis in response to repeated inhalation of acetaldehyde.

METHODS

Subjects

Ten patients with asthma and a mean age of 43.8 years (SEM, 4.4 years) participated in this study (Table I). All were lifelong nonsmokers (never-smokers) and had not had a respiratory tract infection for at least 8 weeks before the study. Each patient with asthma satisfied the American Thoracic Society definition of asthma.¹⁵

This study was carried out when patients' symptoms were mild and stable while they were taking an aerosol β_2 -agonist, oral theophylline, or both. They had not received steroid therapy for at least 8 weeks. This study was approved by the ethics committee of our university hospital.

Study protocol

Bronchoprovocation in response to acetaldehyde was repeated twice, separated by 1 hour, on two occasions 2 weeks apart. Indomethacin, a cyclooxygenase inhibitor, was given orally in a dose of 25 mg four times a day for 3 days and 25 mg at 7:00 AM on the fourth day (test day) in a double-blind, randomized, placebo-controlled, crossover fashion. All medication, except for pretreatment with indomethacin or placebo, was stopped at 10:00 AM on the previous day to allow a washout time of at least 24 hours. The bronchial reactivity to inhaled acetaldehyde was then measured at 10:00 AM on the test day. Informed consent was obtained from all subjects.

Acetaldehyde bronchoprovocation

Acetaldehyde bronchoprovocation was performed by using the method we previously described.^{7,10} Briefly, acetaldehyde was dissolved in physiologic saline solution to make sequential series of doubling concentrations of 80 to 0.16 mg/ml. Saline and acetaldehyde solutions were inhaled from a nebulizer (DeVilbiss 646; DeVilbiss Co., Somerset, Pa.) operated by compressed air at 5 L/min. The nebulizer output was 0.14 ml/min. Saline solution was inhaled first for 2 minutes, and FEV₁ was measured (Chestac 55; Chest Ltd., Nagoya, Japan). If the change in FEV₁ from the baseline value was 10% or less, inhalation of acetaldehyde was started; and if the saline solution caused a change in FEV₁ of more than 10%, the test was stopped or postponed. Acetaldehyde was inhaled for 2 minutes by mouth tidal breathing and followed immediately by measurements of FEV₁. Increasing concentrations of acetaldehyde were inhaled until a fall of 20% or more in FEV₁ occurred. FEV₁ was measured three times, and the best FEV₁ value of three attempts was recorded each time.

Data analysis

The acetaldehyde concentrations producing a 20% fall in FEV₁ (PC₂₀-AcCHO) were calculated from the individual semilogarithmic dose-response curve; the results of PC₂₀-AcCHO were converted to log values for statistical analysis, and values were expressed as geometric standard error of the mean (GSEM). The degree of change in acetaldehyde responsiveness

TABLE I. Subject characteristics

Subject No.	Age (yr)	Sex	FVC (% predicted)	FEV ₁ (% predicted)	FEV ₁ /FVC (%)	Treatment
1	57	F	87.8	71.9	70.0	Sa
2	28	F	119.4	114.4	90.0	Sa
3	28	F	99.4	91.7	81.4	Sa
4	54	M	97.8	86.4	76.1	Sa
5	37	F	118.3	99.3	73.0	Th, Sa
6	57	M	114.9	99.6	76.1	Th, Sa
7	37	M	115.6	98.3	74.2	Sa
8	47	F	102.3	86.4	72.4	Sa
9	65	F	102.5	91.7	73.9	Th, Sa
10	28	M	108.8	92.2	75.8	Sa
Mean	43.8		106.7	93.2	76.4	
SEM	4.4		3.3	3.5	1.9	

FVC, Forced vital capacity; Sa, salbutamol through metered-dose inhaler; Th, orally administered theophylline.

(Δ PC₂₀-AcCHO) was calculated as logarithmic value of the second PC₂₀ minus logarithmic value of the first PC₂₀. Percent change in baseline FEV₁ between the first and second challenges with acetaldehyde (Δ FEV₁) was calculated as: (baseline FEV₁ on second challenge – baseline FEV₁ on first challenge) ÷ baseline FEV₁ on first challenge.

Baseline forced vital capacity and FEV₁ values and Δ FEV₁ were expressed as means and SEM, and these values were compared between placebo and indomethacin by using paired Student's *t* test. After the pretreatment with indomethacin, a 20% fall in FEV₁ was not obtained in one of 10 subjects at the highest concentration of acetaldehyde administered, and therefore a minimal estimate was obtained by calculating the cumulative PC₂₀-AcCHO on the next doubling concentration beyond the highest administered. Because of these censored data, PC₂₀-AcCHO values between placebo and indomethacin treatment were compared by using Wilcoxon signed-rank test. Correlations were obtained by using Spearman's nonparametric rank correlation. Significance was based on a 95% confidence level (*p* < 0.05).

RESULTS

The baseline FEV₁ values before the first and second provocations were 2.82 L (SEM = 0.23) and 2.70 L (SEM = 0.23) after placebo administration and 2.74 L (SEM = 0.22) and 2.67 L (SEM = 0.26) after indomethacin administration. There was no significant difference between baseline FEV₁ values obtained on any two of the four occasions, and Δ FEV₁ values on each test day (–3.8 [SEM = 2.8] after placebo; –3.8 [SEM = 2.3] after indomethacin) were identical.

The geometric mean PC₂₀-AcCHO after placebo administration significantly increased (*p* < 0.01) from 13.0 mg/ml (GSEM = 0.115) to 31.4 mg/ml (GSEM = 0.069) 1 hour after the first challenge (1.26 [SEM = 0.219] doubling doses) (Fig. 1), indicating tachyphylaxis in response to repeated inhalation of acetaldehyde. After indomethacin treatment, no significant increase (a change of 0.490 [GSEM = 0.312] doubling doses) was observed between the first (19.6 mg/ml [GSEM = 0.133]) and second challenges (27.6 mg/ml [GSEM =

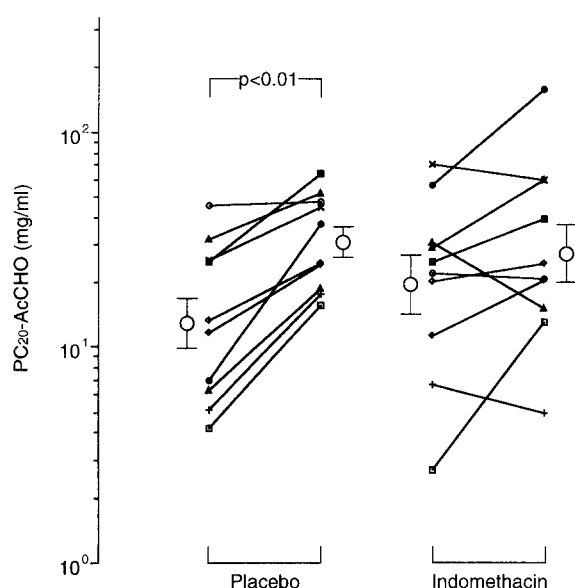


FIG. 1. Effects of indomethacin on tachyphylaxis in response to repeated inhalation of acetaldehyde at 1-hour intervals. Tachyphylactic effect disappeared after pretreatment with indomethacin.

0.94]) (Fig. 1). In addition, there was no significant difference in the first measurements of PC_{20} -AcCHO between indomethacin and placebo. Because the power of the study to determine the difference between the baseline PC_{20} -AcCHO was 71.9%, there is a possibility that the absence of statistical significance is a type 2 error because of the small sample size. The ΔPC_{20} -AcCHO value was significantly ($p < 0.05$) reduced from 0.380 (GSEM = 0.066) to 0.148 (GSEM = 0.094) on pretreatment with indomethacin (Fig. 2). No significant correlation was observed between ΔFEV_1 -AcCHO and ΔPC_{20} -AcCHO on each treatment.

DISCUSSION

In this study we investigated the effect of indomethacin on tachyphylaxis in response to acetaldehyde-induced bronchoconstriction in 10 patients with asthma. The results of this study showed that indomethacin reduced the tachyphylactic effect on repeated inhalation of acetaldehyde at a 1-hour interval and that the degrees of changed bronchial responsiveness to acetaldehyde did not relate to the change in baseline FEV_1 . Because we already reported that inhalation of acetaldehyde by the same method causes no significant bronchoconstriction in healthy nonasthmatic subjects,¹⁰ no control subjects participated in this study.

It has been reported that indomethacin (50 mg, administered four times daily for 4 days) does not affect baseline pulmonary function in patients with asthma.¹⁶ Similar results were obtained in our study, supporting the view that prostaglandins have a minimal effect on the resting tone of the asthmatic airways in vivo. Indomethacin caused no significant change in baseline PC_{20} -

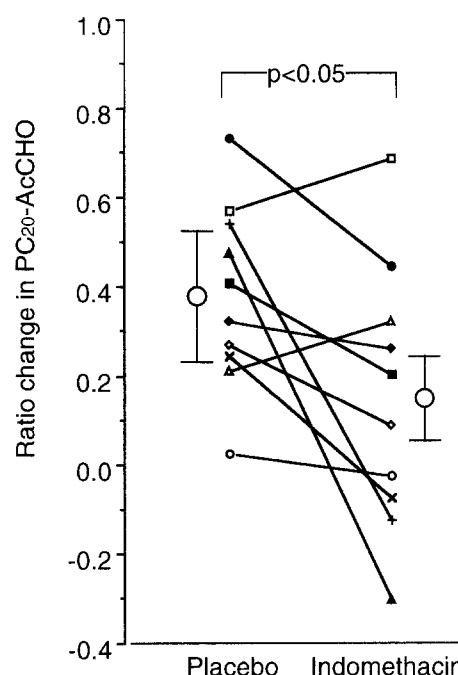


FIG. 2. Degree of tachyphylaxis in response to repeated inhalation of acetaldehyde, expressed as logarithmic value of second PC_{20} minus logarithmic value of first PC_{20} .

AcCHO; this conflicts with our previous finding that a selective thromboxane A_2 synthetase inhibitor, ozagrel hydrochloride, reduced PC_{20} -AcCHO.¹⁴ The possible reasons for this discrepancy are that inhibition of thromboxane synthesis leads to production of bronchodilating prostaglandins^{17, 18} and/or that the dosage of indomethacin used in the study was insufficient to suppress the thromboxane synthesis compared with the dosage of ozagrel hydrochloride in our previous report.¹⁴

In this study there is a slight but not significant tachyphylaxis during indomethacin treatment, suggesting the contribution of bronchodilating prostaglandins, such as prostaglandins E_2 and I_2 , to the tachyphylaxis in response to acetaldehyde. Indeed, a similar phenomenon has been described on bronchoconstriction provoked by exercise,¹⁹ hypertonic saline solution,²⁰ and ultrasonically nebulized distilled water.²¹ We did not examine the effect of thromboxane, one of the cyclooxygenase products, on the tachyphylactic effect in our subjects. Jones et al.²² reported the absence of tachyphylaxis (measured as PC_{20} at a 1-hour interval) in response to repeated inhalation of the thromboxane mimetic U46619, making it likely that the reducing effect of indomethacin on tachyphylaxis in response to acetaldehyde results from a decrease in production of prostaglandins rather than thromboxane.

Our previous report¹⁰ demonstrated that inhaled acetaldehyde produces bronchoconstriction, which is caused by release of histamine in patients with asthma. The origin of

histamine released by acetaldehyde in asthmatic airways is unclear, although acetaldehyde causes dose-dependent histamine release from leukocytes from patients with asthma in vitro.⁸ Prostaglandins E₂ and I₂ are also produced by polymorphonuclear leukocytes,²³ suggesting that acetaldehyde directly produces prostaglandins in asthmatic airways. However, because histamine is a potent inducer of prostaglandin synthesis,²⁴⁻²⁶ histamine released endogenously by acetaldehyde might stimulate the production of prostaglandins.

Because no reducing effect of indomethacin was observed in two subjects (Fig. 2), another mechanism for tachyphylaxis in response to acetaldehyde might be present (e.g., depletion of histamine-releasing cells such as leukocytes, loss of sensitivity to acetaldehyde with regard to histamine release, and release of protective catecholamines). Although the mechanism of the refractory period after an initial exercise task or inhalation of hypertonic saline solution is not related to mediator depletion or release of catecholamines,^{27, 28} further studies of acetaldehyde-induced tachyphylaxis—involving techniques such as the measurement of histamine, prostaglandin, and catecholamine concentrations in the airway or cross-tachyphylaxis—are also required. Furthermore, because there is also concern about nonspecificity of indomethacin for cyclooxygenase products of arachidonic acid metabolism, more specific inhibitors of cyclooxygenase, such as naproxen, would be useful to confirm our conclusion that prostanoids contribute to tachyphylaxis in response to acetaldehyde.

In conclusion, these results suggest that bronchodilating prostaglandins are involved in the tachyphylaxis in response to acetaldehyde-induced bronchoconstriction in patients with asthma.

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