

strong stimulation of PMNs entering the microenvironment of the allergic reaction, of possible great pathogenic importance in IgE-mediated reactions.⁵

REFERENCES

1. Zweiman B, Shalit M, Atkins PC. Release of lactoferrin and elastase in human allergic skin reactions. *J Immunol* 1990;144:3953-60.
2. Sato K, Zweiman B, Moskovitz A, von Allmen C, Atkins PC. Neutrophil activation by skin chamber fluids from allergic reaction sites. *J ALLERGY CLIN IMMUNOL* 1992;90:21-31.
3. Taylor M, Zweiman B, Moskovitz A, von Allmen C, Atkins PC. Platelet activating factor and leukotriene B₄ induced release of lactoferrin from blood neutrophils of atopic and non-atopic individuals. *J ALLERGY CLIN IMMUNOL* 1990;86:740-8.
4. Shalit M, Valone FH, Atkins PC, Ratnoff WD, Goetzl EJ, Zweiman B. Late appearance of phospholipid platelet activating factor and leukotriene D₄ in human skin after repeated antigen challenge. *J ALLERGY CLIN IMMUNOL* 1989;83:691-6.
5. Lemanske RF, Kaliner M. Late-phase IgE-mediated reactions. *J Clin Invest* 1988;81:1-13.

Factors determining bradykinin bronchial responsiveness and refractoriness in asthma

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The mechanism by which bradykinin narrows the airways in human beings is believed to be an indirect one.^{1, 2} In contrast, histamine and methacholine both cause bronchoconstriction directly by stimulating the corresponding receptors on airway smooth muscle. Therefore it is perhaps surprising that three studies²⁻⁴ have shown a correlation in the responsiveness between the two stimuli.

The aim of the present study was to reexamine the relationship between bronchial hyperresponsiveness induced by bradykinin and histamine. We also assessed the incidence and factors that influence the development of refractoriness to bradykinin.

Abbreviations used

FEV ₁	Forced expiratory volume in 1 second
PC ₂₀	Provocative concentration causing a 20% fall in FEV ₁

METHODS

Subjects

Twenty-one nonsmoking, atopic, subjects with mild asthma (13 men) with a mean age of 28.1 years participated in the study. The subjects were chosen without any prior knowledge about their bronchial responsiveness to histamine or bradykinin. The criteria used for selection has been described previously.^{2, 5}

Procedure

The protocols for bronchial challenge with mediators and measurement of forced expiratory volume in 1 second (FEV₁) have been described previously.^{2, 5}

The study consisted of two visits separated by at least 7 days, during which two consecutive concentration-response studies with either inhaled bradykinin or histamine were undertaken in a double-blind, randomized manner. An initial bronchial provocation test with either bradykinin or histamine was performed until FEV₁ fell

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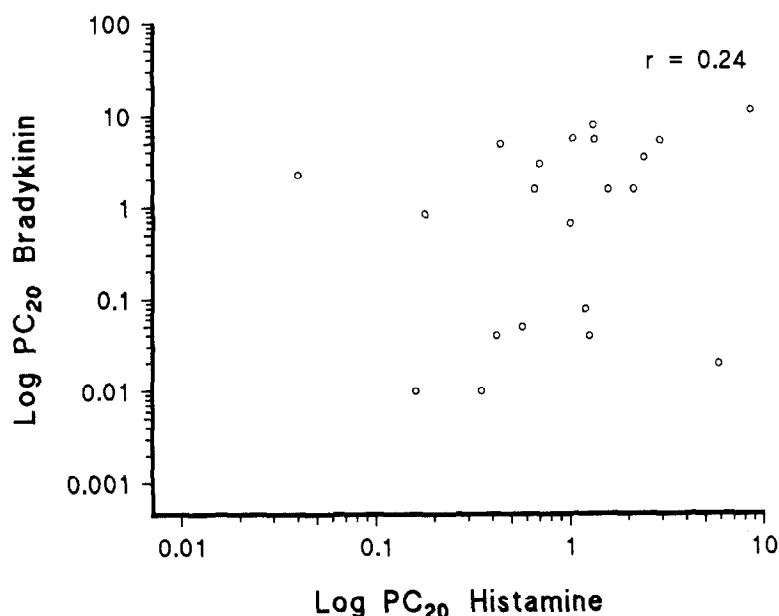


FIG. 1. Correlation between baseline PC₂₀ bradykinin and PC₂₀ histamine values in 21 subjects with asthma.

by more than 20% of baseline value. The subjects were then allowed to recover spontaneously until the FEV₁ had returned to within 5% of baseline value. When this was achieved (1 hour), a second agonist concentration-response challenge was undertaken until FEV₁ was greater than 20% of the original postdiluent value.

Statistical significance was tested by means of two-way analysis of variance and Student's *t* test for paired data. To quantify the loss of agonist responsiveness, an index of refractoriness for bradykinin and histamine was calculated by dividing the second provocative concentration causing a 20% fall in FEV₁ (PC₂₀) by the first PC₂₀, and then the mean of the individual ratios was derived. The relationship between these two indices was assessed by least squares linear regression analysis of the logarithmically transformed values.

RESULTS

Nine of the 21 subjects exhibited refractoriness to repeated inhalation of bradykinin; four of these nine subjects were receiving inhaled beclomethasone therapy. For the group as a whole, the geometric mean PC₂₀ bradykinin increased significantly from 0.63 mg/ml (range, 0.01 to 11.7 mg/ml) after the first challenge to 1.66 mg/ml (range, 0.01 to >16.0 mg/ml) after the second challenge ($p = 0.001$) providing an index of refractoriness of 2.65. The mean PC₂₀ bradykinin after the second challenge was a minimum estimate, because a PC₂₀ was not achieved in two subjects. In contrast, on the histamine study day, the geometric mean PC₂₀ was 0.87 mg/ml (0.04 to 8.41

mg/ml) for the first test and 0.88 mg/ml (0.04 to 6.78 mg/ml) for the second test ($p > 0.05$). The index of refractoriness to bradykinin did not show a significant correlation with baseline histamine reactivity ($r = 0.19$). In contrast, bradykinin refractoriness showed a weak negative correlation with baseline bradykinin reactivity ($r = -0.39$, $p = 0.05$). However, for the group as a whole, no significant correlation could be established between the first PC₂₀ histamine and the first PC₂₀ bradykinin ($p > 0.05$; $r = 0.24$) (Fig. 1).

DISCUSSION

The present study shows, in contrast to previous observations, that tachyphylaxis to bradykinin is not observed in all subjects with asthma. Furthermore, the index of bradykinin refractoriness was not related to baseline bronchial hyperresponsiveness to histamine or bradykinin. In a large group of unselected subjects with asthma we failed to confirm the correlation between bradykinin and histamine reactivity, supporting the view that bradykinin exerts an indirect action on human asthmatic airways.

A close correlation between airway responses to histamine and methacholine has been established, which suggests that common mechanisms regulate this physiologic abnormality. Although a concept that the asthmatic airways are "nonspecifically" hyperresponsive to a wide variety of agonist has been developed, this has not been

found to be the case when either histamine or methacholine are compared with other stimuli such as adenosine 5'-monophosphate, platelet activating factor, neurokinin A, exercise, sulfur dioxide or sodium metabisulfite, hypertonic saline, and distilled water.

The term "indirect bronchial hyperresponsiveness" has been suggested for those stimuli that provoke bronchoconstriction through release of pharmacologically active mediators. Evidence that the group of indirect stimuli reveal a different component of airway responsiveness than do the more "conventional" stimuli such as methacholine and histamine has been provided by the observation that topical corticosteroids can decrease responsiveness to these stimuli but not to sulfur dioxide. On the basis of the present evidence it seems unlikely that bradykinin has a predominant action by direct constriction of airway smooth muscle. It follows from this that indices of bradykinin and histamine responsiveness are unlikely to correlate. However, at least

three studies have shown a correlation between these two indices,²⁻⁴ but the subject groups were small ($n < 10$) and were selected.

REFERENCES

1. Simonsson BG, Skoogh BE, Bergh NP, Andersson BR, Svedmyr N. In vivo and in vitro effect of bradykinin on bronchial motor tone in normal subjects and patients with airways obstruction. *Respiration* 1973;30:378-88.
2. Fuller RW, Dixon CMS, Cuss FMC, Barnes PJ. Bradykinin-induced bronchoconstriction in humans: mode of action. *Am Rev Respir Dis* 1987;135:176-80.
3. Polosa R, Holgate ST. Comparative airway response to inhaled bradykinin, kallidin and [des-arg⁹]-bradykinin in normal and asthmatic subjects. *Am Rev Respir Dis* 1990; 142:1367-71.
4. Polosa R, Phillips GD, Lai CKW, Holgate ST. Contribution of histamine and prostanoids to bronchoconstriction provoked by inhaled bradykinin in atopic asthma. *Allergy* 1990;45:174-82.
5. Polosa R, Lai CKW, Robinson C, Holgate ST. The influence of cyclooxygenase inhibition on the loss of bronchoconstrictor response to repeated bradykinin challenge in asthma. *Eur Respir J* 1990;3:914-21.