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Effect of Ipratropium Bromide on Bronchospasm Induced by Inhalation of Hypotonic Solutions

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ABSTRACT

Pérez Neria, José, Luis Manuel Montaño R., Gilberto Monge L., Horacio Rubio M., and Moisés Selman L.: Effect of ipratropium bromide on bronchospasm induced by inhalation of hypotonic solutions. Drug Dev. Res. **11**:117–121, 1987.

In order to determine the role of the cholinergic system in the bronchoconstriction induced by hypotonic solutions, 30 subjects were studied: 15 asthmatic patients and 15 healthy volunteers to whom distilled water was administered by inhalation through ultrasonic nebulization. All asthmatic patients showed a 20% reduction of forced expiratory volume in 1 sec in relation to the initial value. Bronchospasm was not detected in any of the healthy subjects. After 24 hr, ipratropium bromide in aerosol was administered 1 hr before distilled water nebulization, and this prevented the bronchoconstriction response in 11 of the 15 asthmatic patients. These results suggest that such bronchospasm is mediated at least in part by vagal reflex.

Key words: water-induced bronchospasm

INTRODUCTION

Asthma is characterized by a variable and reversible airway obstruction and an exaggerated bronchoconstriction response to a variety of stimuli [Liker, 1982; Barnes, 1983]. This response can be measured in the laboratory by exposing the subject to situations that induce bronchospasm. These methods are widely known as bronchial provocation tests and can be specific or nonspecific [Pepys and Hutchcroft, 1975], and both have demonstrated its usefulness in the etiologic diagnosis and analysis of asthma. Specific inhalation tests include immu-

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nologic challenge with specific antigens [Permutt et al., 1977]. Provocation tests of nonspecific responsiveness are usually performed with pharmacologic agents such as methacoline or histamine [Hargreave et al., 1981] or with response to physical stimuli such as exercise [Anderson, 1983].

On the other hand, it has been demonstrated that the inhalation of hypotonic solutions through ultrasonic nebulization is a sensitive, inexpensive, and useful nonspecific test that reveals the presence of bronchial hyperreactivity [Cheney and Butler, 1968; Allegra and Bianco, 1980; Pérez Neria et al., 1985].

Although the precipitating mechanism of bronchospasm by inhalation of hypotonic solutions is not known, several theories of its pathogenesis have been proposed. One of them suggested that inhalation of such solutions induces water loss from the respiratory mucosa leading to bronchial hyperreactivity [Elwood et al., 1982; Hahn et al., 1984]. On the other hand, other authors have reported that hypotonic solution induces the release of histamine from mast cells [Cheney and Butler, 1968; Schoeffel et al., 1981], which suggests a different mechanism.

The aim of the present study was to determine the possible role of the cholinergic system in the bronchospasm induced by inhalation of hypotonic solutions.

PATIENTS AND METHODS

Fifteen asthmatic patients and 15 healthy subjects of both sexes were studied. Exclusion criteria included diabetes, arterial hypertension, cardiovascular disease, respiratory infections, and being under steroid treatment.

In the asthmatic group, the age ranged from 17 to 58 years, with a mean of 29.3 $\pm$ 4.3 years; there were four males and 11 females. The established diagnosis was "mixed" asthma, that is, induced by both immunologic and nonimmunologic stimuli. The duration of the disease varied from 2 to 28 years, with an average of 7 $\pm$ 1.8 years. In the control group, the age ranged from 20 to 35 years with a mean of 26.6 $\pm$ 1.2 years; there were ten males and five females.

Distilled water was administered by aerosol, through a De Vilbis Ultrasonic Nebulizer (Model 35B). Nebulization output was 6 ml/min. Inhalation was performed through a plastic mask with lateral holes of 2 cm in diameter connected to a polyvinyl tube 30 cm in length and 1.9 cm in diameter. The subjects studied did not receive any medication for 12 hr before the study.

In order to evaluate the presence of bronchospasm, the 1-sec forced expiratory volume (FEV_1) was measured with a Neumoscan S-301 Spirometer. After a baseline spirometry determination, distilled water was administered by inhalation to the subjects. FEV_1 determinations were done every minute. The inhalation of water was continuous until a 20% or greater reduction in FEV_1 was observed. This volume represented the PC_{20}. The inhaled dose of distilled water was calculated multiplying 6 ml/min by the elapsed time. If after 15 min a 20% reduction in FEV_1 was not observed, then the test was stopped.

The same procedure was repeated 24 hr later, but the patients and controls received single doses of 40 μg (two puffs) of inhaled ipratropium bromide 1 hr before the challenge with distilled water.

Statistical analysis was done using the paired and non-paired Student t-test. P-values of .05 or less were considered to be statistically significant. Values are reported in the text as mean $\pm$ SEM.

Drug

Ipratropium bromide in aerosol was the drug used in the experiment (Lab. Boehringer Ingelheim).

RESULTS

Inhalation of distilled water in the asthmatic patients induced bronchospasm of the central airways as shown in Table 1. FEV_1 was increased after inhalation of distilled water and such difference was statistically significant ($P < .05$). The percentage change in FEV_1 from baseline was -33.27 ± 2.61. The changes in FEV_1 in all the asthmatic patients were 20% or higher. In relation to the time of inhalation, none of the asthmatic patients completed 15 min (Table 1).

After inhalation of ipratropium bromide, 11 of the 15 patients (73.3%) tolerated 15 min of nebulization distilled water (90 ml). These results compared to the ones obtained without ipratropium bromide showed a difference that was stastistically significant ($P < .05$) (Table1).

On the other hand, a statistically significant difference ($P < .001$) was observed in the percentage change in FEV_1 induced by the hypotonic solution before and after administration of ipratropium bromide. In other words, premedication with this cholinergic antagonist prevented the bronchoconstriction response of the central airways to the inhalation of distilled water (Fig. 1).

In the control group there was no statistically significant difference in FEV_1 under baseline conditions and after inhalation of distilled water (Table 2). The percentage change in FEV_1 from baseline was $+13.4 \pm 5.48$, a positive difference in contrast to the negative difference found in the asthmatic group (Fig. 1).

The percentage change in FEV_1 from baseline in male patients after nebulization distilled water was -35.39 ± 5.31 and in females it was -33.21 ± 3.08. After premedication of ipratropium bromide these values were -5.04 ± 6.46 and -12.96 ± 4.97, respectively. In both groups there were no statistically significant differences.

DISCUSSION

Our data suggest that the ipratropium bromide pretreatment attenuates the bronchoconstriction response elicited by inhalation of hypotonic solutions in 11 of the 15 asthmatic subjects.

The mechanism through which these hypotonic solutions induce bronchospasm has not been elucidated. However, it has been proposed that the inhalation of such solutions can lead, owing to changes in osmolarity, to water loss from the respiratory mucosa and, in the long run, to an increase in the bronchial hyperreactivity [Elwood et al., 1982; Hahn et al., 1984].

On the other hand, it has been demonstrated that there is a small population of luminal mast cells in the airways of asthmatic patients that release chemical mediators to different stimuli including hypotonic solutions [Schoeffel et al., 1981; Barnes, 1983]. Such released mediators, like histamine, increase the permeability of the airway, which would lead to an increased exposure of the surface irritant receptors [Hulbert et al., 1981; Hogg, 1982]. Once these epithelial receptors are exposed, they are stimulated by the histamine released, evoking a bronchoconstrictor response through a vagal reflex [Gold et al., 1972; Dixon et al., 1979].

TABLE 1. Results of FEV_1 Time and Volume Before and After Inhalation of Distilled Water and After Administration of Ipratropium Bromide (IB) in the Asthmatic Group

	FEV_1 (liters/sec)		Time of inhalation	Inhaled volume
$\bar{X} \pm$ SEM	Baseline	After inhalation	(min)	of water (ml)
Distilled water	1.37 ± 0.12	0.91 ± 0.08	2.73 ± 0.41	16.4 ± 2.45
	($P < .05$)			
Distilled water plus IB	1.46 ± 0.16	1.34 ± 0.14	11.87 ± 1.37	71.2 ± 8.49
	(NS)		($P < .05$)	($P < .05$)

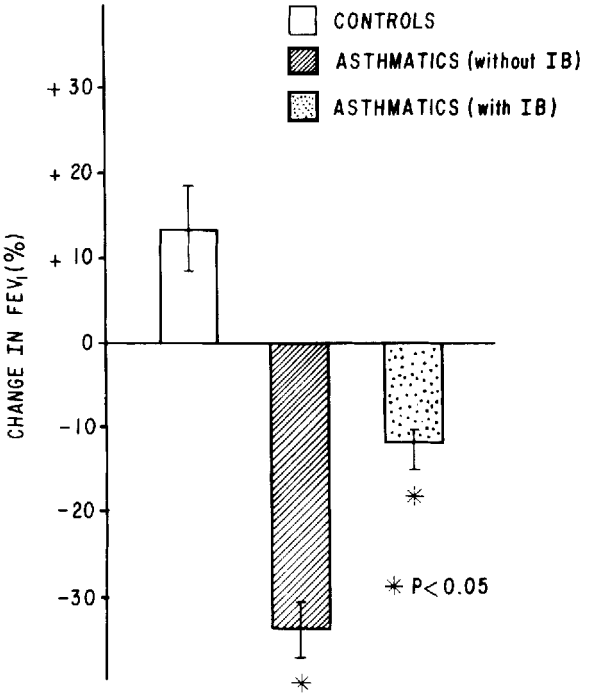

Fig. 1. The percentage change in FEV_1 before and after inhalation of distilled water in the control group and in the asthmatic group before and after inhalation of distilled water alone and plus ipratropium bromide (IB).

TABLE 2. Results of FEV_1 Time and Volume Determinations Before and After Inhalation of Distilled Water in the Control Group

	FEV_1 (liters/sec)		Time of inhalation (min)	Inhaled volume of water (ml)
	Baseline	After inhalation		
$\bar{X} \pm$ SEM	2.23 ± 0.17	2.47 ± 0.15	15 ± 0	90 ± 0

In the same context, another possible explanation of the bronchospasm induced by distilled water could be the permanent damage of the bronchial epithelium in asthmatic subjects [Hogg, 1982], which would lead to a greater exposition of the surface irritant receptors, and in this way the simple inhalation of a hypotonic solution would be enough to stimulate such receptors and induce bronchospasm by a vagal reflex.

In addition, there is evidence that mast cells have cholinergic receptors on their surface [Kaliner, 1977]. In this way, it is possible that some mediators released by mast cells could be partially responsible for this bronchospasm. The prevention of water-induced bronchoconstriction by disodium cromoglycate suggests that biochemical mediator release may be involved in this reaction [Orr and Cox, 1969].

Other alternative hypotheses could be that airway cooling by room temperature aerosol or that the irritant effects of the aerosol particles of distilled water are responsible for bronchoconstriction, but these do not appear to be valid because ultrasonic aerosols of isotonic saline at room temperature do not cause bronchoconstriction [Eschenbacher et al., 1984].

Nevertheless, the fact that in our study ipratropium bromide clearly decreases the bronchospasm produced by distilled water, strongly suggests that this is due at least in part to

a vagal reflex, probably by activation of subepithelial afferent neural receptors that cause vagally mediated bronchoconstriction.

However, it should be emphasized that this cholinergic antagonist did not revert the bronchospasm in 26.7% of the asthmatic patients, suggesting that other noncholinergic mechanisms are involved in the pathogenesis of this condition. Although one of them could be the production of bioactive substances by the mast cells, another one may be the recently discovered non-cholinergic-mediated contractile system, which is reported to use substance P as its mediator [Grundstrom and Anderson, 1985].

REFERENCES

Allegra, L. and Bianco, S.: Non-specific broncho-reactivity obtained with ultrasonic aerosol of distilled water. Eur. J. Respir. Dis. **61** (Suppl.):41–49, 1980.

Anderson, S.D.: Current concepts of exercise induced asthma. Allergy **38:**289–302, 1983.

Barnes, J.P.: Pathogenesis of asthma: A review. J.R. Soc. Med. **76:**580–586, 1983.

Cheney, F.W., and Butler, J.: The effect of ultrasonically produced aerosols on airway resistance in man. Anesthesiology **29:**1099–1106, 1968.

Dixon, M., Jackson, D.M., and Richards, I.M.: The effect of H_1-and H_2-receptor agonist and antagonist on total lung resistance, dynamic lung compliance and irritant receptor discharge in the anesthetized dog. Br. J. Pharmacol. **66:**203–209, 1979.

Elwood, R.K., Hogg, J.C., and Pare, P.D.: Airway response to osmolar challenge in asthma. (Abstract) Am. Rev. Respir. Dis. **125:**61, 1982.

Eschenbacher, W.L., Boushey, H.A., and Sheppard, D.: Alteration in osmolarity of inhaled aerosols causes bronchoconstriction and cough, but absence of a permeant union causes cough alone. Am. Rev. Respir. Dis. **129:**211–215, 1984.

Gold, W.M., Kessler, G.F., and Yu, D.Y.C.: Role of vagus nerves in experimental asthma in allergic dogs. J. Appl. Physiol. **33:**719–725, 1972.

Grundstrom, N., and Anderson, R.G.G.: In vivo demonstration of alpha-2-adrenoceptor-mediated inhibition of the excitatory non-cholinergic neurotransmission in guinea pig airways. Naunyn Schmiedebergs Arch. Pharmacol. **328:**236–240, 1985.

Hahn, A., Anderson, S.D., Morton, A.R., Black, J.L., and Fitch, K.D.: A reinterpretation of the effect of temperature and water content of the inspired air exercise-induced asthma. Am. Rev. Respir. Dis. **130:**575–579, 1984.

Hargreave, F.E., Ryan, G., Thomson, N.C., O'Byrne, P.M., Latimer, K., and Juniper, E.F.: Bronchial responsiveness to histamine or metacholine in asthma: Measurement and clinical significance. J. Allergy Clin. Immunol. **68:**347–355, 1981.

Hulbert, W.C., Walker, D.C., Jackson, A., and Hogg, J.C.: Airway permeability to horseradish peroxidase in guinea pigs: The repair phase after injury by cigarette smoke. Am. Rev. Respir. Dis. **123:**320–326, 1981.

Hogg, J.C.: The pathophysiology of asthma. Chest **1:**85–125, 1982.

Kaliner, M.: Human lung tissue and anaphylaxis: The role of cyclic GMP as a modulator of the immunologically induced secretory process. J. Allergy Clin. Immunol. **60:**204–211, 1977.

Lilker, E.S.: Asthma is a disease: A new theory of the pathogenesis. Chest **82:**263–264, 1982.

Orr, T.S.C., and Cox, J.S.G.: Disodium cromoglycate, an inhibitor of mast cell degranulation and histamine release induced by phospholipase A. Nature **233:**197–198, 1969.

Pepys, J., and Hutchcroft, B.J.: Bronchial provocation tests in etiologic diagnosis and analysis of asthma. Am. Rev. Respir. Dis. **112:**829–859, 1975.

Pérez Neria, J., Ocaña, H., Rojas, A., Rubio, H., Carrillo, G., and Selman, M.: Obstrucción bronquial por inhalación de agua destilada. Arch. Bronconeumol. **21:**269–272, 1985.

Permutt, S., Rosenthal, R.R., Norman, P.S., and Menkes, H.A.: Bronchial challenge in ragweed-sensitive patients. In Lichtenstein, L.M. and Austen, K.E. (eds): "Asthma. Physiology, Immunopharmacology and Treatment." New York: Academic Press, pp., 265–282, 1977.

Schoeffel, R.E., Anderson, S.D., and Altounyan, R.E.C.: Bronchial hyperreactivity in response to inhalation of ultrasonically nebulized solutions of distilled water and saline. 3r Med. J. [Clin. Res.] **283:**1285–1287, 1981.