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ORIGINAL ARTICLE

Nitric oxide in children with persistent asthma

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Abstract

Objective: To assess the difference in exhaled nitric oxide levels in atopic and nonatopic asthmatic patients treated with anti-inflammatory drugs, and to compare exhaled nitric oxide measurement with lung function tests.

Methods: Cross-sectional study with 45 consecutively selected patients with moderate and severe persistent asthma, aged between 6 and 17 years, and treated with anti-inflammatory drugs for at least 1 year. The patients were split into two groups: atopic ones (with positive skin tests) and nonatopic ones. The clinical and functional assessments and the measurement of exhaled nitric oxide were carried out concomitantly.

Results: There was a male predominance (62.5%), with an age range between 6 and 13 years (mean of 10.4 years) in 85% of the patients. Neither the symptoms associated with asthma ($p = 0.07$), allergic rhinitis ($p = 0.17$), food allergy ($p = 0.09$), necessity of systemic corticosteroids ($p = 0.10$), antileukotrienes ($p = 0.20$) and antihistamines ($p = 0.70$), nor the three parameters used to assess lung function (FEV_1, FEV_1/FVC and $FEF_{25-75\%}$, $p \geq 0.14$) were statistically significant. The frequency of eczema ($p < 0.005$) and exhaled nitric oxide levels ($p < 0.001$) were higher among atopic patients.

Conclusion: Results suggest that clinical and functional stability of asthma among atopic patients does not necessarily reflect an efficient control over the inflammatory process and a higher probability for recurrence after discontinuation of anti-inflammatory therapy.

J Pediatr (Rio J). 2006;82(3):193-6: Asthma, atopy, exhaled nitric oxide.

Introduction

Chronic airway inflammation plays a key role in asthma, but no marker has been routinely used for this condition in pediatric practice. Nitric oxide (NO) is a marker of this inflammation. Exhaled nitric oxide level is a useful indicator, and can be determined instantly (on-line) through sensitive, practical, easy, and noninvasive methods, which provide immediate, reliable, and standardized results.[1,2] Measurement of exhaled nitric oxide can be used for the diagnosis and monitoring of responses to anti-inflammatory therapy, since untreated patients show high levels of exhaled nitric oxide. Due to the wide clinical and functional variability of this disorder, treatment schemes often have to be adjusted.[3-7]

Exhaled nitric oxide can help with the detection of subclinical disorders, of insufficient anti-inflammatory therapy, and of treatment compliance, in addition to allowing the identification of patients at risk for asthma exacerbation.[8] Inhaled or systemic corticosteroids reduce exhaled nitric oxide levels.[9,10] Thus, exhaled nitric oxide measurement can be used to monitor and quantify inflammation and it is therefore useful for individual control, and especially, for the management of inhaled corticosteroid doses.

The aim of the present study is to determine the clinical and functional profile and exhaled nitric oxide levels in atopic and nonatopic children and adolescents with persistent asthma.

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Methods

Patients and place of study

This was a cross-sectional study conducted at the Division of Pediatric Pulmonology and Allergology of Hospital Necker-Enfants Malades (Paris, France), with 45 consecutively selected asthmatic children.

Inclusion and exclusion criteria

Patients with moderate and severe persistent asthma, selected according to the Global Initiative for Asthma,[11] submitted to inhaled corticosteroid therapy for at least 12 months, were included in the study. Children with mild intermittent or persistent asthma who did not receive inhaled corticosteroids and who had other underlying disorders were excluded from the study.

Analyzed groups

Children and adolescents were categorized into atopic and nonatopic. Atopy was defined as the presence of positive skin test results.

Exhaled nitric oxide measurement

Exhaled nitric oxide was measured on-line, using Endono® (Seres, Aix-en-Provence, France), with the child in a comfortably sitting position, without physical activity prior to examination and without the use of any medication on the day of examination. Exhaled nitric oxide is measured by chemiluminescence, which counts the photons emitted at the return of NO molecules produced by NO reaction in the presence of excess ozone (O_3) to a more stable state; the light signal is proportional to the NO concentration.

NO was measured during a prolonged expiration of at least six seconds, in order to reach a plateau of at least three seconds, at a flow of 50 ml/s.[12] A plateau of two seconds in children younger than 12 years reduces the reliability of the exam.[13]

The available devices for this exam detect NO of bronchopulmonary origin in the minimum values of a part per billion (ppb).[14] The cutoff point for the upper limit of normal corresponded to 10 ppb, according to the specificity of Endono® and according to Franklin,[15] and was also the reference value adopted by the Division where the study was carried out.

Exhaled nitric oxide levels vary with devices and manufacturers, usually ranging between 10 and 20 ppb. However, in healthy children and adolescents, levels around 10 ppb, ranging from 7.2 ppb (6.4-8.0 ppb)[15] to 8.7 ppb (8.1-9.2 ppb) have been described in the literature.[16] These levels are elevated in asthmatic patients, with rates 1.6 to 4.4 times greater than those found in healthy individuals.[17] During treatment with inhaled or systemic corticosteroids, these levels decrease or even return to normal.

Statistical analysis

Frequency distribution and means were calculated, and the chi-square test, Fisher's exact test, and Student's t test were used.

Results

Of 45 children and adolescents, 24 (53.3%) were atopic and 21 (46.6%), nonatopic. The characteristics of the study population are shown next (Table 1).

There was a male predominance (62.5%), and the age range was 6-13 years in 85% of the patients. The classification of asthma severity was similar in both groups ($p = 1.0$), but the group of atopic patients revealed a higher incidence of eczema ($p = 0.005$). No statistical significance was observed for the presence of symptoms ($p = 0.07$), allergic rhinitis ($p = 0.17$), food allergy ($p = 0.09$), greater necessity of systemic corticosteroids ($p = 0.10$), antileukotrienes ($p = 0.20$) and antihistamines ($p = 0.70$).

All patients received inhaled corticosteroids; in the first group, the dose of inhaled corticosteroids averaged 396 µg (± SD 78.0) and, in the other group, it averaged 264.3 µg (± SD 80.0) ($p = 0.002$).

Table 2 shows the lung function test results, with parameters expressed in percentage predicted values and exhaled nitric oxide levels.

Even though there was no statistical significance for the three parameters used to assess lung function (FEV_1, FEV_1/FVC and $FEF_{25-75\%}$), exhaled nitric oxide measurements showed a statistically significant difference (16.7 ppb and 5.3 ppb) between the groups ($p < 0.01$). Atopic patients had an average exhaled nitric oxide level 3.1 times higher, thus showing the persistence of inflammatory activity despite the use of anti-inflammatory drugs.

Seven patients, six atopic and one nonatopic, received systemic corticosteroids (Table 1). Although exhaled nitric oxide is recommended for severe persistent asthma, its level among atopic patients was remarkably higher (respectively, 16.5 ppb, 64.0 ppb, 31.5 ppb, 16.0 ppb 16.0 ppb and 16.0 ppb, mean of 26.6 ppb) than that obtained for the nonatopic patient (5.3 ppb, $p < 0.001$).

Discussion

The average exhaled nitric oxide levels (5.3 ppb) observed among nonatopic patients after treatment with inhaled corticosteroids, antileukotrienes and antihistamines are consistent with the reference values.[15-17]

There was no agreement as to whether the presence of symptoms was higher in the atopic patient. In the present study, no statistically significant difference was observed between the groups; however, some authors have described such significance.[18,19]

The present study revealed that, even though asthma in both groups had been treated with inhaled corticosteroids and/or antileukotrienes and/or antihistamines and/or systemic corticosteroids, atopic asthmatic children showed higher exhaled nitric oxide levels, compared to nonatopic asthmatic children ($p < 0.001$), which is in agreement with the studies on the behavior of exhaled nitric oxide in cases of atopy. By assessing 235 children aged between 8 and 14 years, Leuppi et al.[20] found an exhaled nitric oxide level of 18.6 ppb in atopic patients and of 12.9 ppb in nonatopic ones ($p = 0.001$). A similar study, conducted by Broxai et al.[21] with 429 children, showed a geometric mean of 10 ppb for exhaled nitric oxide in atopic individuals and of 7.7 ppb in nonatopic ones ($p < 0.01$). Likewise, Silvestri et al.[22] investigated 112 atopic and nonatopic children with moderate and severe persistent asthma and mean

Table 1 - Characteristics of the study population

Variable	Atopic (n = 24)		Nonatopic (n = 21)		p
	n	%	n	%	
Gender					
Male	15	62.5	14	66.7	
Female	9	37.5	7	33.3	0.98
Age (years)					
6-10	10	41.7	13	61.9	
11-13	10	41.7	6	28.6	
14-17	4	16.6	2	9.5	0.29
Asthma severity					
Moderate	2	8.3	2	9.6	
Severe	22	91.7	19	90.4	1.0
Symptoms					0.07
Yes	13	54.2	5	23.8	
No	11	45.8	16	76.2	
Allergic rhinitis					
Yes	8	33.3	3	14.3	
No	16	66.7	18	85.7	0.17
Eczema					
Yes	6	25.0	zero	0.0	
No	18	75.0	21	100.0	0.005
Food allergy					
Yes	2	8.3	zero	0.0	
No	22	91.7	21	100.0	0.09
Corresponding dose of beclomethasone					
Up to 800 µg	20	83.3	20	95.2	
> 800 µg	4	16.7	1	4.8	0.35
Use of antileukotrienes					
Yes	9	37.5	4	19.0	
No	15	62.5	17	81.0	0.20
Use of antihistamines					
Yes	5	20.8	3	14.3	
No	19	79.2	18	85.7	0.70
Use of systemic corticosteroids					
Yes	6	25.0	1	4.7	
No	18	75.0	20	95.3	0.10

Table 2 - Spirometry and fractional exhaled nitric oxide measurement results in atopic and nonatopic asthmatic patients

	Atopic (n = 24)	Nonatopic (n = 21)	p
FEV_1 (% of the predicted value)	99.1	95.1	0.23
FEV_1/FVC (% of the predicted value)	95.8	92.0	0.14
FEF_{25-75} (% of the predicted value)	82.5	80.6	0.5
FeNO (ppb)	16.7	5.3	< 0.001

FEF_{25-75} = forced expiratory flow 25-75%; FeNO = fractional exhaled nitric oxide in parts per billion; FEV_1 = forced expiratory volume in one second; FEV_1/FVC = forced expiratory volume in one second /forced vital capacity.

age of 10.9 years, and found an exhaled nitric oxide level of 23.9 ppb in atopic individuals and of 7.6 ppb in nonatopic ones (p = 0.0001). These results suggest that atopy itself induces the production of different cell populations and/or the production of different cytokines, regardless of the use of anti-inflammatory drugs. These authors admitted that the decrease in airway pH causes bronchospasm, production of proinflammatory substances and conversion of nitrogen dioxide to NO. It should be highlighted that, similarly to the present study, Silvestri et al. did not find any correlation between exhaled nitric oxide and pulmonary parameters (p > 0.1).[22]

Results are especially interesting when lung function is assessed, since its parameters did not show any correlation with exhaled nitric oxide, demonstrating that normal spirometry does not reflect the absence of an ongoing inflammatory process and that it is not possible to affirm that asthma in a given patient is under control based on normal test values.

Persistently high exhaled nitric oxide levels may be due to the lack of control of the inflammation. In this study, it was persistently high in six atopic children treated with systemic corticosteroids (26.6 ppb). Buchvald et al.[23] raised the hypothesis that there may be individuals with different responses of exhaled nitric oxide to corticosteroids.

Cross-sectional studies are limited, but they can provide information for possible changes, for the reduction or increase in the dose of inhaled corticosteroids in the prophylaxis of asthma. This is what the study conducted by Smith et al.[24] showed. They investigated 46 patients aged between 12 and 75 years, using exhaled nitric oxide concentration to adjust the dose of inhaled corticosteroids. By the end of the observation period, they noted that this group used 45% less inhaled corticosteroid than the control group with 48 patients treated according to international consensus criteria. The dose of fluticasone corresponded to 370 µg and 641 µg, respectively (p = 0.003). Exacerbation was higher in the control group (p = 0.23), with an exhaled nitric oxide level of 6.4 ppb. The authors concluded that the control group received a higher dose of inhaled corticosteroids than necessary and suggested that exhaled nitric oxide is a logical alternative to the adjustment of inhaled corticosteroid therapy. Pijnenburg et al.[25] found that some atopic patients continue to have high levels of exhaled nitric oxide even after the prolonged use of inhaled corticosteroids, and that these levels remain high with the correct use of the inhalation technique and after an increase in the dose of inhaled corticosteroids.

Finally, the present study showed that exhaled nitric oxide measurement was better than spirometry, and the presence of symptoms for assessment of atopic asthmatic patients and the results obtained suggest that these patients are seemingly more susceptible to relapses than nonatopic individuals.

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