Original Article

Correlation of Atopy and FeNO in Allergic Rhinitis: An Indian Study

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ABSTRACT

Background. Fractional exhaled nitric oxide (FeNO) is a non-invasive marker of airway inflammation. Limited published data are available on the effect of atopy on FeNO in allergic rhinitis.

Objectives. To investigate the relationship between atopy and FeNO in patients with allergic rhinitis.

Methods. Patients with allergic rhinitis were assessed for atopy and exhaled breath analysis of nitric oxide. Atopy was assessed by skin prick testing (SPT) against 58 common aero-allergens; a wheal size of 3mm or more as compared to buffer saline was considered positive. Patients were labelled to be atopic if they had at least one positive SPT result. The measurement of FeNO level was done by using NIOX chemiluminescence analyser.

Results. Forty-nine participants (26 males) aged between 8-50 years were studied and 31 of them were found to be atopic. The average value of FeNO in the subjects studied (n=49) was 26.0±22.7 parts per billion (ppb) with significantly higher values being observed in atopic group as compared to non-atopic group (34.2±24.3 *versus* 11.9±9.0 ppb; p<0 .05).

Conclusion. As FeNO is a marker of lower airway inflammation, significantly higher FeNO levels in atopic allergic rhinitis patients suggest that it may be a predictor for onset of asthma in these patients.

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Key words: Fractional exhaled nitric oxide, Allergic rhinitis, Atopy, Skin prick testing.

INTRODUCTION

Allergic sensitisation in an individual can manifest as bronchial asthma, rhinitis and is sometimes independent of symptoms. This sensitisation leads to accumulation of inflammatory cells which release various inflammatory mediators in the airways.[1] Eosinophilic airway inflammation has been demonstrated by invasive methods like eosinophilia in bronchoalveolar lavage, as well as by non-invasive method, like induced sputum eosinophilia. These established markers of airway inflammation have excellent correlation with the level of fractional exhaled nitric oxide (FeNO).[2,3] Hence, FeNO has been validated as a marker for eosinophilic inflammation of airways.[4] The concept of 'one airway, one disease' is now well recognised on the basis of epidemiological, pathophysiological, and clinical data.[5,6] The FeNO level has been found to be raised in patients of allergic rhinitis.[7]

After an extensive search of databases and *to the best of our knowledge* no study correlating the marker of airway inflammation, namely, FeNO and atopy was found in Indian population. Hence, the present study was undertaken to study this correlation.

MATERIAL AND METHODS

Study Population and Design

The study was conducted at the out-patient department (OPD) of the Vallabhbhai Patel Chest Institute, Delhi between September 2011 to November 2011. Patients with allergic rhinitis visiting the OPD were included for the study. The study group consisted of 49 patients. The diagnosis of allergic rhinitis was made based on the clinical definition of allergic rhinitis as per Allergic Rhinitis and Its Impact on Asthma (ARIA) guidelines (2008 update).[8] Patients with symptoms of rhinorrhoea, nasal obstruction, nasal itching and sneezing which were reversible spontaneously or with treatment were diagnosed to have allergic rhinitis.[8] Patients with

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history of wheezing, breathlessness, having received asthma medication or demonstrating obstructive pattern on pulmonary function testing were excluded from the study. Other exclusion criteria were: (i) smokers, defined as persons who have smoked 100 cigarettes or more in their entire lifetime or currently smoking any number of cigarettes;[9] (ii) nasal/oral steroid intake in the preceding one month; (iii) occurrence of recent upper or lower respiratory tract infections; and (iv) history of urticaria or eczema.

Skin prick testing (SPT) to common aeroallergens was performed in all patients as per standard guidelines,[10] 58 aeroallergens which are routinely used for allergy testing. If indicated immunotherapy was administered subsequently. To perform the SPT, a small drop of test reagent (allergen extract) was placed on the surface of the forearm. Then a disposable hypodermic needle (26G) with its bevel facing up was passed through the drop and inserted into the skin about 1mm at a low angle. The needle tip was gently lifted upwards a bit without inducing bleeding and the needle withdrawn slowly. After about two minutes, the drop was gently wiped off with dry cotton. The test reading was done after 15 to 20 minutes. Atopy was defined as a positive SPT (wheal diameter of >3 mm as compared to buffer saline as control) for at least one aeroallergen.[10] The patient with negative SPT to all aeroallergens was labelled as non-atopic.

Exhaled Nitric Oxide

FeNO measurements were performed by chemiluminescence analyser (NIOX Aerocrine AB, Solna, Sweden) in accordance with the 2005 American Thoracic Society/ European Respiratory Society (ATS/ERS) recommendations for standardised online measurements.[11] In this technique the patient was asked to insert the mouthpiece, inhale for 2 to 3 seconds through mouth to total lung capacity (TLC) and then exhale immediately. Patient exhaled at a constant flow rate (50 mL/s) from TLC to residual volume without breath-holding. The duration of exhalation had to be sufficient (>4 seconds in children younger than 12 years and >6 seconds in children older than 12 years) and the concentration of nitric oxide (NO) was evaluated over a 3-second window. Repeated, reproducible exhalations were performed to obtain at least two NO plateau values that agreed within 10% of each other. The mean level of two reproducible recordings was used as the result value.

Statistical Analysis

Data analysis was performed using Statistical Package for the Social Sciences (version 14.0 for windows; SPSS, Chicago, IL, USA). The univariate analyses of factors associated with FeNO was done using Pearson's correlation. The measurements for FeNO were compared between atopic and non-atopic groups using the independent sample t-test.

RESULTS

The study included 49 patients (26 males). Their average age was 23 years (range 8 to 50 years). The characteristics of study population are shown in table 1. SPT was performed in all the patients and 31 patients were found to be atopic. Out of 31 SPT positive atopic patients, 26 had SPT results positive against two or more aeroallergen, three patients had positive results against for two aeroallergens and two patients had SPT positive for a single aeroallergen. The remaining 18 patients who did not have a positive SPT result were labelled as non-atopic.

Table 1. Characteristics of the study subjects (n=49) and univariate analysis of factors associated with FeNO

Characteristics	Mean±SD	r Value	p Value
Age (years)	23.2±9.8	0.303	0.024
Anthropometric Measurements			
Height (cm)	153.5±13.8	0.148	0.309
Weight (kg)	52.6±13.1	0.139	0.342
Body mass index (kg/m²)	22.1±3.9	0.084	0.564
Duration of disease	5.7±5.1	0.097	0.508
Pulmonary Function Test			
FVC % predicted	91.8±11.9	0.066	0.652
FEV_1 % predicted	90.7±13.5	–0.051	0.727
FEV_1 /FVC % predicted	95.5±7.9	–0.103	0.481

FVC= Forced vital capacity; FEV_1=Forced expiratory volume in one second; FEV_1 /FVC %=Ratio of forced expiratory volume in one second to forced vital capacity.

The FeNO levels in all patients ranged from 3ppb to 107 ppb; the geometric mean FeNO level in study population was 26.0±22.7 ppb. The mean level of FeNO in atopic group was 34.2±24.3 ppb and mean level of FeNO in non-atopic group was 11.9±9.0 ppb (Table 2). The patients in atopic group had higher mean FeNO levels when compared to patients of non-atopic groups and the difference was statistically significant ($p<0.005$). The FeNO levels significantly and positively correlated to age ($r=0.303$, $p<0.05$; Table 1). However, anthropometric variables like height, weight and body mass index (BMI) kg/m^2 did not show statistically significant correlation to FeNO (Table 1).

Further for the purpose of analysis, the patients were divided into four groups based on SPT result analysis, namely, Group I: non-atopics, Group II: 1-5 SPT positives; Group III: 6-10 SPT positives; and Group IV: 11-24 SPT positives. The mean FeNO value was higher in Group III followed by Group II and then Group IV. Group I (SPT negative) had lowest mean FeNO levels (Table 3).

Table 2. Characteristics of atopic and non-atopic study groups

Continuous Variable	Atopic* (n=31)	Non-atopic* (n=18)	p value
Age (years)	24.1±1.9	21.7±8.4	0.417
Anthropometric Measurements			
Height (cm)	154.1±14.8	152.4±12.0	0.387
Weight (kg)	52.4±13.3	53.1±13.0	0.866
Body mass index (kg/m^2)	21.8±4.2	22.5±3.5	0.599
Duration of disease	6.6±5.8	4.2±3.3	0.133
FeNO (ppb)	34.2±24.3	11.9±9.0	0.002
Pulmonary Function Test			
FVC % predicted	92.5±13.2	90.6±9.7	0.591
FEV_1 % predicted	91.0±15.4	90.1±9.7	0.816
FEV_1/FVC % predicted	94.5±8.8	97.1±6.1	0.281
FEF_{25-75}% predicted (%)	89.3±26.2	92.9±23.5	0.625

* Data are expressed as mean±SD; FVC=Forced vital capacity; FEV_1=Forced expiratory volume in one second; FEV_1/FVC%=Ratio of forced expiratory volume in one second to forced vital capacity; FEF_{25-75}%=Average forced expiratory flow rate over the middle 50% of the FVC

The FeNO levels in male patients (25.5±22.0 ppb) were lower as compared to levels of female patients (26.6±23.9 ppb), though the difference was statistically not significant. In our study, the pulmonary function parameters like forced expiratory volume in one second (FEV_1), forced vital capacity (FVC), ratio of forced expiratory volume in one second to forced vital capacity (FEV_1/FVC) and average forced expiratory flow rate over the middle 50% of the FVC (FEF_{25-75}) did not show any significant correlation with the levels of FeNO.

Table 3. Group classification on the basis of number of aeroallergens with positive SPT result

Group	Number of Aeroallergens with Positive SPT	Number of Patients	FeNO (Mean±SD)
I	0	18	11.9±9.0
II	1-5	12	30.7±13.2
III	6-10	12	40.3±34.1
IV	11-24	7	29.7±19.2

SPT=Skin prick test

DISCUSSION

Nitric oxide is produced by enzyme nitric oxide synthase expressed in many cells of the upper and lower respiratory system, such as ciliated epithelial cells, mucosal cells and endothelial cells.[12] FeNO measurement is non-invasive, simple and well tolerated method and with the availability of standardised instruments and guidelines it has become an important tool for research and as a clinical biomarker to assess airway inflammation.[4,11] FeNO has been found to be an important adjunct in diagnosis and management of asthma.[13] The FeNO level measurement has been evaluated in various studies and standardised for the diagnosis or to support the diagnosis in cases of eosinophilic inflammation of airways, bronchial hyperreactivity, asthma and also for corticosteroid responsiveness.[4] In our study of allergic rhinitis patients, atopic and non-atopic groups did not have any significant difference in baseline variables (Table 2). However, the patients in atopic group had a higher level of FeNO when compared to the patients in non-atopic group ($p<0.005$). This suggests that atopic patients of allergic rhinitis may also have asymptomatic lower

airway inflammation consistent with unified allergic airway hypothesis. Our finding of correlation of atopic status and significant increase in FeNO levels are consistent with the results of previous studies.[14-16] Jouaville *et al*[17] studied the levels of FeNO in atopic and non-atopic rhinitis and concluded that atopic rhinitis patients have higher levels of FeNO than non-atopic patients of rhinitis (28.2±9.5 ppb *vs* 22.5±17.2 ppb). Similarly Alvarez *et al*[18] also reported the increased levels of FeNO in atopic subjects of allergic rhinitis in the absence of lower airway symptoms. They suggested it to be because of the presence of subclinical inflammation of lower airways. This was further evaluated with biopsy finding of eosioniphil accumulation in upper and lower airways even in the absence of asthma symptoms. Some studies[19,20] have reported increased levels of FeNO in atopic patients, irrespective of presence of respiratory symptoms. However, another study[21] did not report any significant differences in levels of FeNO in atopy or rhinitis.

Also, there have been studies[16,22,23] which have found positive correlation between the number of SPT positives and the levels of FeNO in asthmatic patients. Hence, FeNO levels may be a surrogate marker for atopic status. In the present study, when FeNO levels were analysed after classifying on the basis of number of SPT positives, it was observed that with increase in number of SPT positives there is an increase in FeNO levels except in Group IV (Table 3). This could be due to less number of patients in Group IV. However, the difference in FeNO levels in different groups was not statistically significant.

The results of our study suggested an age dependent increase of FeNO levels, which has been similar to previously reported in one study.[24] There have been conflicting reports regarding the role of gender influencing the levels of FeNO with some studies suggesting males exhaling higher concentration as compared to females[25] while other disagree.[26] We found that females having higher FeNO levels than males, though the results were statistically not significant.

To the best of our knowledge and database search, study correlating FeNO and atopy in allergic rhinitis patients has never been done in India. In our study we found that the atopic status of the patients significantly correlates with the levels of FeNO. Hence, increased levels of FeNO may be an indicator of allergic sensitisation (atopy). These atopic allergic rhinitis patients did not have symptoms of asthma, but had increased FeNO levels (compared to non-atopic allergic rhinitis). We hypothesise that these may be having subclinical inflammation of lower airways which may lead to the development of asthma in future. This needs a further large scale longitudinal study.

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