

Mite avoidance can reduce air trapping and airway inflammation in allergic asthmatic children

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Summary

Background We investigated the effects of prolonged allergen avoidance in 18 house dust mite-sensitized asthmatic children during a prolonged residential period at a high altitude, allergen-free environment.

Methods Evaluations of residual volume (RV) and exhaled nitric oxide (eNO) were performed (i) at admission to the residential house in September, (ii) in December after 3 months of stay, (iii) in January after 15 days at home, exposed to allergens, and (iv) in June after 9 months of stay.

Results During the study period RV showed a significant decrease in December (from $117.5 \pm 7.7\%$ to $96.5 \pm 3.2\%$) ($P < 0.02$) and a following increase in January ($126.2 \pm 17.2\%$), after allergen re-exposure ($P < 0.03$). RV decreased again in June at the end of the study period ($91.1 \pm 6.0\%$) ($P = 0.001$). FEV₁, FEF_{25–75} and VC values did not present significant variations. ENO showed a significant decrease in December after 3 months at high altitude (from 21.3 ± 3.9 p.p.b. to 11.9 ± 1.7 p.p.b.) ($P = 0.03$), but no further significant change. No correlation was found between lung volumes and eNO, probably reflecting different aspects of asthma.

Conclusions Results suggest that RV may be more sensitive than other respiratory function parameters in identifying children with air trapping, being influenced significantly as the inflammatory indices by effective allergen avoidance/exposure regimen.

Keywords allergen avoidance, exhaled nitric oxide, residual volume

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Introduction

The exposure to offending allergens in sensitized asthmatic patients causes eosinophil influx and epithelial damage of the airway, which may lead to an increase in bronchial inflammation, to airway remodelling and to bronchial hyper-reactivity (BHR) [1, 2]. A reduced forced expiratory volume in 1 s (FEV₁) to vital capacity (VC) ratio is indicative of airflow limitation and may distinguish obstructive from non-obstructive patterns [3], but it cannot indicate the precise localization of airway obstruction, particularly when the small airways are involved. There are functional patterns characterized by a concomitant decrease in FEV₁ and VC and with a consequent normal FEV₁ to VC ratio. In these conditions the increase of residual volume (RV) values may reflect the obstructive impairment of the small airways [4]. The obstructive syndrome localized at the small airway level is characterized by the increase of RV and RV/total lung capacity (TLC) ratio and it is a feature of a phenomenon of air trapping, which may be present even in asymptomatic asthma [4]. In asthmatic patients with normal or near normal flow-volume values increased RV has been

demonstrated to be associated with severe exacerbations of asthma [5].

We have previously reported the reduction of allergen-induced BHR by effective allergen avoidance at high altitude [6]. The prolonged stay at high altitude, allergen-free environment, leads to a reduction of symptoms, an increase of spirometric values [7] and a reduction of bronchial inflammatory indices [8]. Recently, we utilized eNO as sensitive markers of effective allergen avoidance [9]. In the present paper we studied the changes in RV and lung volumes in asthmatic house dust mite (HDM)-sensitized children, while resident for a long period in an environment free of the offending allergen. We investigated whether not only the anti-inflammatory therapy but also the effective allergen avoidance could ameliorate the pattern of air trapping and whether this phenomenon could be accompanied by a parallel reduction of eNO as index of airway inflammation.

Methods

Patients

Eighteen children (mean age $\pm$ SD, 10.7 ± 1.6 years) were evaluated. All children had moderate to severe asthma according to the definition of the American Thoracic Society [10] and the minimum medication required to achieve adequate control [11]. All patients were sensitive to HDM,

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Dermatophagoides pteronyssinus and *farinae*, as demonstrated by positive skin prick tests and positive RAST (score > 3) for the allergens. They were resident in the Istituto Pio XII, Misurina, at 1756 m above sea level, where the routine analysis for HDM always failed to reveal the presence of mite-allergen. The domestic levels of HDM were evaluated for each child who participated in the study.

Study design

The children stayed at high altitude in Misurina for the whole scholastic year, 9 months, from September to June. The children went home for 15 days for the Christmas holidays, being therefore exposed to the offending allergen HDM, from December to January. The lung volume measurements and the eNO evaluations were performed in four periods [1]: in September, within 3 days of first admission to the institute [2], in December, after 3 months of stay, and within 2 days before leaving the residential house for the Christmas holidays [3], in January, at the return to the institute after 15 days at home, and [4] in June, after 9 months at high altitude in Misurina.

Patients' treatments

At the admission to the residential house in September the patients were in regular treatment with anti-inflammatory drugs (Table 1); within a few weeks of admission the regular therapies were gradually withdrawn due to symptomatic improvement. During the Christmas holidays the children returned home, receiving preventive regular treatment with inhaled steroids (Table 1) that was continued for a further week after the return to the institute in January and then gradually withdrawn.

Lung function tests

The lung volume measurements were obtained using a water-sealed lung volume spirometer, which uses a washout system with removal of CO₂, and O₂ delivery control system and

analysis of the helium dilution by a fast response cell (Baires, Biorevo mod., Biomedin, Padova, Italy). Appropriate corrections for temperature and gas viscosity were applied [12]. The tests were always performed at the same time of the day.

ENO measurements

Exhaled NO was measured using a chemiluminescence analyser (Logan LR 2149, Rochester, Kent, UK). Briefly, the children were asked to perform a single slow exhalation through a mouthpiece against a resistance. A biofeedback device maintained a 5–6 L/min steady expiratory flow, assuring the closure of the soft palate and separating the nasopharynx from the oropharynx. ENO values were registered at the plateau of the end-exhaled reading according to the guidelines [13].

Measurement of mite allergen levels

Parents were asked to collect dust from the mattresses of the patients by using a vacuum cleaner with a new bag and vacuuming the mattress surface over an approximate area of 2 m² for 5 min. The parents were well instructed in the procedure as a part of the educational programme that is regularly carried out at the residential house. The dust was then stored at 4 °C until the process for measurement of mite group I allergen levels, which was performed with monoclonal antibodies in a two site immunometric ELISA [14].

Statistical analysis

The study hypothesis was tested by examining internal changes of RV, FEV₁, VC, functional residual capacity (FRC) and forced expiratory flow between 25% and 75% of the vital capacity (FEF_{25–75}) and eNO during the study period. Comparison for contiguous times of the exposure-avoidance study was planned *a priori* and the analysis of variance (ANOVA) was used for comparing observations. Correlations between the levels of eNO and the values of RV, FEV₁ and FEF_{25–75} were tested by Spearman's correlation test.

Table 1. Individual data for demographics and treatment at each time of evaluation. M = male, F = female, Flu = fluticasone (doses are expressed as µg/day), Bud = budesonide (doses are expressed as µg/day). For all time points β₂ agonists were allowed as needed for all subjects

Patient N°	Gender	Age (yr)	Height (cm)	Weight (kg)	Treatment September	Treatment December	Treatment January	Treatment June
1	M	11	143	41	Flu 300	–	Flu 300	–
2	M	13	162	55	Flu 200	–	Flu 300	–
3	M	13	156	48	Flu 600	–	Flu 300	–
4	M	11	151	53	Flu 200	–	Flu 300	–
5	M	7	124	20	Bud 400	–	Flu 300	–
6	M	10	137	37	Bud 400	–	Flu 300	–
7	M	11	136	34	Bud 400	–	Flu 300	–
8	M	9	130	33	Flu 200	–	Flu 300	–
9	F	9	129	33	Flu 400	–	Flu 300	–
10	M	13	168	63	Bud 400	–	Flu 300	–
11	M	12	147	37	Bud 400	–	Flu 300	–
12	M	11	140	36	Flu 300	–	Flu 300	–
13	M	9	146	53	Flu 200	–	Flu 300	–
14	M	10	136	27	Flu 300	–	Flu 300	–
15	M	12	150	42	Flu 300	–	Flu 300	–
16	M	10	146	40	Bud 400	–	Flu 300	–
17	M	11	159	60	Flu 200	–	Flu 300	–
18	M	10	123	24	Flu 300	–	Flu 300	–

The local hospital ethical committee approved the study and the children and their parents gave informed consents.

Results

The results obtained, expressed as group mean $\pm$ standard error of the estimate ($\pm$ SEM), are summarized in Table 2, where the *P*-value is equal to comparison for contiguous times. Table 3 and Table 4 describe the single patient's pulmonary parameters for each of the four times of the study. During the study period the RV values (Fig. 1) showed a significant decrease ($P < 0.02$) after 3 months of stay at high altitude, from $117.5 \pm 7.7\%$ in September to $96.5 \pm 3.2\%$ in December. In January, when the children returned to the residential house, after 15 days in the presence of HDM at home, there was an increase of RV ($126.2 \pm 17.2\%$) that was significant in comparison to December ($P < 0.03$). The RV decreased again and in June

was $91.1 \pm 6.0\%$, being significantly different from January ($P = 0.001$).

The RV to TLC ratio (Table 2) presented a significant decrease between September and December ($P = 0.003$) and a following significant increase between December and January ($P = 0.002$). In 6 months of residency in altitude the RV to TLC ratio showed a decrease that was significantly different from January ($P = 0.002$). The evaluation of the FRC (Table 2) showed again a significant decrease from September to December ($P = 0.016$); in January we observed an increase that failed to be significant in comparison to December. In June there was a decrease of FRC that was significantly different from the values observed in January ($P = 0.012$). When the variations of FEV₁, FEF₂₅₋₇₅ and VC were considered, no significant difference between periods was observed (Table 2) (Fig. 1). ENO presented a significant decrease from September (21.3 ± 3.9 p.p.b.) to December (11.9 ± 1.7 p.p.b.) ($P = 0.03$) (Fig. 1). No further significant

Table 2. Respiratory parameters at different times. RV = residual volume, TLC = total lung capacity, FRC = functional residual capacity, FEV₁ = forced expiratory volume in 1 s, FEF₂₅₋₇₅ = forced expiratory flow between 25% and 75% of the vital capacity, VC = vital capacity. *P*-value is equal to comparison for contiguous times. NS = non-significant

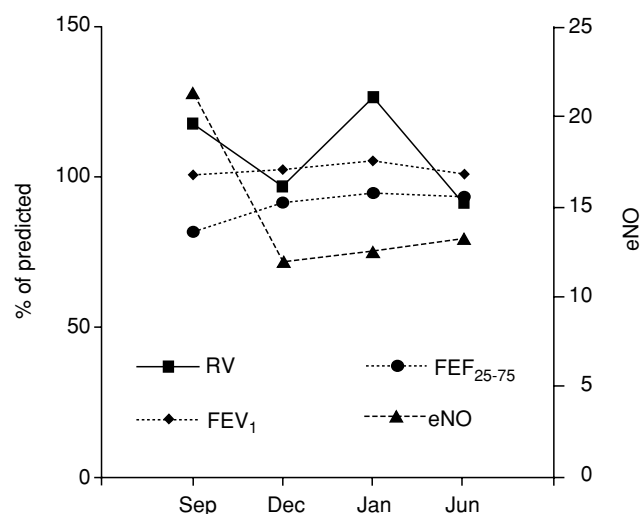
	September	December	January	June
RV (%)	117.7 $\pm$ 7.7	96.5 $\pm$ 3.2 <i>P</i> < 0.02	126.2 $\pm$ 17.2 <i>P</i> < 0.03	91.1 $\pm$ 6.0 <i>P</i> = 0.001
RV/TLC (%)	107.8 $\pm$ 5.3	93.7 $\pm$ 4.8 <i>P</i> = 0.003	107.3 $\pm$ 4.5 <i>P</i> = 0.002	90.2 $\pm$ 5.1 <i>P</i> = 0.002
FRC (%)	100.2 $\pm$ 4.4	89.6 $\pm$ 3.0 <i>P</i> = 0.016	99.8 $\pm$ 5.5 NS	89.3 $\pm$ 3.6 0.001
FEV ₁ (%)	100.5 $\pm$ 3.6	102.0 $\pm$ 2.9 NS	105.1 $\pm$ 2.6 NS	100.5 $\pm$ 2.3 NS
FEF ₂₅₋₇₅ (%)	81.5 $\pm$ 5.9	91.3 $\pm$ 5.1 NS	94.4 $\pm$ 5.8 NS	93.2 $\pm$ 5.9 NS
VC (%)	109.8 $\pm$ 5.8	108.5 $\pm$ 5.3 NS	117.3 $\pm$ 5.9 NS	107.6 $\pm$ 4.9 NS

Table 3. Single patient's pulmonary parameters at different times. Time 1 = September, Time 2 = December, Time 3 = January, Time 4 = June

Patient number	1 VC (L)	2 VC (L)	3 VC (L)	4 VC (L)	1 FEV ₁ (L)	2 FEV ₁ (L)	3 FEV ₁ (L)	4 FEV ₁ (L)	1 FEF ₂₅₋₇₅ (L/s)	2 FEF ₂₅₋₇₅ (L/s)	3 FEF ₂₅₋₇₅ (L/s)	4 FEF ₂₅₋₇₅ (L/s)
1	2.82	2.91	3.04	3.1	2.27	2.3	2.54	2.48	2.22	2.14	2.53	2.25
2	4.73	4.98	4.77	5.16	3.55	3.88	3.7	3.9	2.91	3.41	3.27	3.2
3	2.8	3.56	3.55	3.75	1.48	3.06	2.77	2.64	0.64	3.09	2.2	1.86
4	3.36	3.27	3.36	3.71	2.83	2.75	2.93	3.07	2.93	2.95	3.56	3.14
5	1.63	1.75	1.76	1.75	1.57	1.55	1.55	1.54	2.51	2.72	2.64	2.82
6	2.68	2.83	3.01	2.96	2.04	2.32	2.4	2.35	1.7	2.32	2.26	2.02
7	2.59	2.63	2.58	2.74	2.03	1.99	1.91	2.03	1.76	1.99	1.49	1.61
8	2.34	2.2	2.29	2.47	1.87	1.8	1.87	2.02	1.77	1.77	1.85	2.03
9	1.97	2.09	2.19	2.29	1.75	1.81	1.98	2.0	2.34	2.03	2.61	2.41
10	4.05	4.42	4.67	4.37	2.52	3.06	3.38	4.08	1.5	3.4	2.57	4.96
11	3.09	3.2	3.29	3.29	2.33	2.34	2.55	2.53	1.87	1.82	2.19	2.14
12	2.63	2.56	2.49	2.61	2.35	2.42	2.27	2.33	2.81	3.25	3.03	2.67
13	2.59	2.71	2.71	2.76	2.16	2.22	2.38	2.51	2.29	2.29	3.19	3.57
14	2.12	2.17	2.24	2.19	1.91	1.92	2	2.02	2.22	2.24	2.41	2.57
15	3.19	3.22	3.46	3.46	2.66	2.94	2.93	3.16	2.67	3.72	3.06	3.96
16		2.24	2.5	2.52		1.48	1.93	2.15		0.9	1.77	2.38
17	3.49	3.42		3.57	2.81	2.59		2.85	2.44	2.15		2.45
18	2.21	2.29		2.27	1.72	1.73		1.73	1.51	1.41		1.49

Table 4. Single patient's pulmonary parameters at different times. Time 1 = September, Time 2 = December, Time 3 = January, Time 4 = June

Patient number	1 RV (L)	2 RV (L)	3 RV (L)	4 RV (L)	1 TLC (L)	2 TLC (L)	3 TLC (L)	4 TLC (L)	1 RV/TLC (%)	2 RV/TLC (%)	3 RV/TLC (%)	4 RV/TLC (%)
1	0.55	0.59	0.56	0.45	3.37	3.50	3.60	3.55	16.37	16.76	15.65	12.67
2	0.82	0.86	1.06	0.9	5.55	5.83	5.83	6.06	14.84	14.67	18.18	14.85
3	0.65	0.77	2.2	0.87	3.45	4.33	5.75	4.62	18.74	17.79	38.26	18.84
4	0.65	0.6	0.46	0.51	4.00	3.87	3.83	4.23	16.16	15.51	12.03	12.17
5	0.67	0.46	0.47	0.44	2.30	2.22	2.23	2.19	29.13	20.85	21.13	20.25
6	0.92	0.66	1.13	0.57	3.61	3.49	4.14	3.53	25.58	18.95	27.22	16.16
7	0.71	0.69		0.92	3.30	3.32		3.66	21.52	20.79	44.06	25.02
8	0.78	0.72	0.54	0.49	3.12	2.92	2.83	2.97	25.07	24.76	19.13	16.64
9	0.47	0.46	0.16	0.31	2.44	2.55	2.34	2.60	19.19	18.14	6.07	11.99
10	1.57	1.14	1.54	1.4	5.62	5.56	6.20	5.77	27.91	20.48	24.77	24.27
11	0.86	0.78	0.8	0.74	3.96	3.99	4.09	4.03	21.83	19.64	19.52	18.37
12	0.85	0.76	0.88	0.77	3.48	3.32	3.37	3.38	24.45	22.77	26.23	22.84
13	1.05	0.77	0.91	0.68	3.64	3.48	3.62	3.45	28.92	22.02	25.21	19.83
14	0.89	0.6	0.75	0.61	3.01	2.76	2.99	2.8	29.59	21.56	25.17	21.76
15	0.78	0.97	1.09	1.08	3.97	4.19	4.55	4.54	19.65	23.07	23.95	23.84
16		0.66	0.77	0.5		2.90	3.06	3.02		22.71	12.67	16.47
17	0.94	0.86		1	4.43	4.28		4.57	21.18	20.11		21.84
18	0.94	0.58		0.72	3.15	2.88		2.99	29.77	20.24		24

**Fig. 1.** Variations of different parameters for each time point of the study. The results are expressed as mean $\pm$ standard error of the estimate (SEM). RV = residual volume, FEV₁ = forced expiratory volume in 1 second, FEV_{25–75} = forced expiratory flow between 25% and 75% of the vital capacity, eNO = exhaled nitric oxide.

change in January (12.5 ± 2.6 p.p.b.) and in June (13.2 ± 2.0 p.p.b.) was observed.

No significant correlation was found between eNO and the lung volumes. The measurement (mean $\pm$ SD) of mite allergen levels, expressed in micrograms of mite group I allergens for gram of dust, was 23.9 ± 5.5 .

Discussion

In our study population the presence at home of HDM group I allergens was associated with significant high values of RV and eNO. The decrease in RV was significant in December, after 3 months of stay at high altitude, in an environment absolutely

free of mites, and it was maintained by the end of the study period in June. The levels of eNO, a sensitive marker of airway inflammation, showed a parallel significant decrease after 3 months of allergen avoidance, demonstrating that also airway inflammation was significantly related to the avoidance regimen, as previously described [8, 9]. When the children returned home for 15 days, in the presence of the offending allergens, the inhaled steroid treatment was able to prevent the increase in eNO and deterioration of other lung function parameters, i.e. FEV₁ and FEV_{25–75}, but it did not prevent peripheral airway obstruction and the associated increase of RV.

We failed to show any correlation between eNO and the lung function measurements. According to previous observations this lack of correlation may suggest that these measures are reflective of different aspects of asthma [15, 16]. eNO has been proposed to be a more sensitive marker of deterioration of asthma than either symptoms or lung function and to be affected by inhaled steroid treatment [17]. However, data from our study demonstrate that RV is even more sensitive than eNO as a marker of allergen exposure/avoidance and it is less influenced by the inhaled steroid treatment. Our results confirm previous observations [4] that in asthmatics there could be an increase in RV and in RV/TLC ratio even when the FEV₁ and VC are within the normal limit of predicted values. This condition should be considered a feature of air trapping and it was defined as small airway obstruction syndrome [4].

Residual volume changes have been studied using different bronchial challenges, such as methacoline and allergen, which produced at the early asthmatic response the same changes in flows and volumes, suggesting that, though different mechanisms, the site of obstruction was similar [18]. In the case of the late asthmatic response, artificially induced by allergen challenges, the increase of RV was much greater, reflecting the involvement of more peripheral bronchial walls and parenchyma with a mechanism that might be similar to the situation of the children when they arrived at high altitude [19, 20]. Air trapping may produce alveolar hyperinflation that could be the

only baseline pulmonary abnormality in asthmatics during long symptom-free intervals [21, 22].

It has been observed that chronic airway obstruction and hyperinflation may result in increased total lung capacity in adults who have suffered asthma from childhood in comparison with subjects who developed asthma after the age of 18 years [23]. The lung hyperinflation may represent one of the most important causes of dyspnoea and the response to bronchodilator therapy is due to the fact that the consequent increase in bronchial caliber allows the patient to breathe at a lower lung volume [24]. Furthermore, in asthmatic children the presence of hyperinflation, even if with normal or near normal FEV₁ and with the disease apparently under control, could be associated with more severe asthma exacerbations, characterized clinically by life-threatening episodes [5]. The group of asthmatic children with hyperinflation presented significant decreased lung elasticity in comparison with normal subjects [25], a greater incidence of previous respiratory failure and a higher incidence of steroid requirement [5]. For this reason the loss of elastic recoil, i.e. hyperinflation, appears to represent a marker for severe, life-threatening asthma in children.

Debate exists on the opportunity to perform not only spirometric but also total lung volume measurements, that in most cases are considered time-consuming and expensive [26]. In our study when the children first arrived at high altitude in September, the RV measurements and the eNO levels showed significant high values despite the fact that all the children were on inhaled steroids and their asthma was well controlled. As previously indicated, asthma control is often inappropriately used to define asthma severity [11]: our study population presented a well-controlled asthma, being on steroid treatment. Despite asthma control being achieved, the RV and eNO high levels for September confirmed that our patients presented indeed moderate to severe asthma.

Our results support the use of lung volume measurements and the evaluation of RV at least in moderate to severe asthmatic children also when there is a normal FEV₁/VC ratio. RV values may be significantly elevated even in apparently well-controlled asthmatics, secondary to inflammation, goblet cell metaplasia and/or narrowing due to collagen deposition and peribronchial fibrosis that may be present in the distal airways of asthmatics [2, 27–29]. These modifications, probably present even in early asthma and in childhood [30], may be sufficient to cause an increase of expiratory flow limitation and airway closure and, if unresolved during the early phase of the disease, to lead to the loss of elastic recoil characteristic of severe asthma.

In conclusion, in the present study at high altitude the prolonged allergen avoidance led to an increase of the airway caliber and to a parallel reduction of inflammation with a significant decrease, respectively, of RV and eNO. These data further support the importance of an effective allergen-avoidance programme in the treatment of HDM-sensitive asthmatic children. Because the presence of air trapping may represent a major risk factor for severe, life-threatening asthma in children, it is recommended to verify the lung volumes in moderate to severe asthmatics. The changes in lung volumes should be particularly evaluated when assessing the effectiveness of a long-term anti-inflammatory therapy or of an allergen avoidance programme.

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