

ORIGINAL PAPER

Respiratory symptoms, bronchial hyper-responsiveness, and eosinophilic airway inflammation in patients with moderate-to-severe atopic dermatitis

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Summary

Background Patients with atopic dermatitis (AD) often have symptoms suggestive of asthma or rhinitis. The prevalence and signs of respiratory disease in AD patients have been studied to a limited extent.

Objectives To assess the prevalence and clustering of respiratory symptoms, bronchial hyper-responsiveness (BHR), and eosinophilic airway inflammation in patients with moderate-to-severe AD.

Methods Eighty-six consecutive patients with moderate-to-severe AD and 49 randomly selected control subjects without AD were studied by questionnaire, flow volume spirometry, histamine challenge to detect BHR, induced sputum test to detect eosinophilic airway inflammation, and skin prick tests (SPTs) and total serum immunoglobulin (Ig)E measurements to detect atopy.

Results The patients with AD showed increased risk of physician-diagnosed asthma (36% vs. 2%, odds ratio (OR) 10.1, confidence interval (CI) 1.3–79.7, $P = 0.03$), physician-diagnosed allergic rhinitis (AR) (45% vs. 6%, OR 4.5, CI 1.2–16.7, $P = 0.02$), BHR (51% vs. 10%, OR 5.5, CI 1.5–20.1, $P = 0.01$), and sputum eosinophilia (81% vs. 11%, OR 76.1, CI 9.3–623.5, $P < 0.0001$) compared with the control subjects. In AD patients, elevated s-IgE and positive SPTs were associated with the occurrence of physician-diagnosed asthma and AR, BHR, and the presence of sputum eosinophilia.

Conclusions BHR and eosinophilic airway inflammation are more common in patients with AD than in control subjects. The highest prevalences were seen in patients with AD who were SPT positive and had high IgE levels. Longitudinal studies are needed to assess the outcome of patients with signs of airway disease, in order to identify those who need early initiation of asthma treatment.

Keywords allergic rhinitis, asthma, atopic dermatitis, bronchial hyper-responsiveness, serum immunoglobulin E, skin prick tests, sputum eosinophilia

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Introduction

Atopic dermatitis (AD), allergic rhinitis (AR), and allergic asthma frequently occur in the same patient. The diseases share many common predisposing factors, and they frequently appear in a consecutive manner, which has been called the atopic march. The development of asthma is often preceded by AD, which usually appears in early childhood, to be followed by AR and asthma in pre-school or school age or early adulthood.

Chronic airway inflammation with eosinophils and bronchial hyper-responsiveness (BHR) represent the main

pathophysiological changes in asthma [1, 2]. BHR has been observed to occur in AD patients, even in the absence of an asthma diagnosis [3–5]. However, studies on the prevalence of BHR in AD patients are few, and in even fewer cases have control groups been included. Atopy determined with positive skin prick tests (SPTs) has been shown to be associated with signs of airway inflammation [6–9]. However, it is not clear how prevalent eosinophilic airway inflammation is in patients with AD and with what probability the above risk factors predict the appearance of asthma. BHR and eosinophilic airway inflammation

could be warning signs that should lead the clinician to consider early initiation of anti-inflammatory asthma treatment [10, 11]. A clinically feasible test to investigate eosinophilic airway inflammation in AD patients would be the induced sputum test, which is non-invasive and safe [12–14]. In this cross-sectional study, we investigated the prevalence of respiratory symptoms, BHR, and sputum eosinophilia in patients with moderate-to-severe AD and in a group of randomly selected control subjects without AD or apparent other atopic diseases. The dependence of airway disease on AD severity, SPT positivity, and serum immunoglobulin E (s-IgE) level was also examined.

Methods

Subjects

A total of 86 consecutive outpatients older than 13 years with moderate-to-severe AD were recruited at the Skin and Allergy Hospital, Helsinki University Central Hospital. For inclusion, the patients were required to fulfil Hanifin–Rajka criteria [15] for atopic eczema, and have over 5% body surface area affected with eczema. The severity of eczema was evaluated by Rajka–Langeland criteria [16] and the body surface area affected with eczema (mean 29%, range 6–100%). The eczema was severe in 24 (28%) patients and all of them had 36% or more of the body surface affected.

Except for possible asthma or AR, the patients were free from other significant chronic diseases. The main treatment for AD had been topical corticosteroids. Additionally, 13 patients had used cyclosporin A, four had used occasional short courses of peroral corticosteroids, and thirteen patients had undergone UV phototherapy during the preceding year. A randomly selected control group included 49 volunteer medical students and staff members of similar age as the patients, but with no AD or apparent other atopic diseases. All subjects gave their written informed consent, and the study was approved by the institutional ethics committee. Medical histories were collected from all subjects, with particular reference to asthma and AR and to the manner in which these diseases had been treated. Subject characteristics are summarized in Table 1.

AD patients' s-IgE was assayed. Observed levels were regarded as elevated if they exceeded the following age-related reference range upper limits: over 17 years 110 kU/L, 15–17 years 160 kU/L, and 13–14 years 320 kU/L.

All subjects underwent SPTs with 10 mg/mL histamine dihydrochloride as the positive control, solvent as the negative control, and ten allergens: birch, timothy, mugwort, cat, dog, cow, horse, *Cladosporium herbarum*, *Dermatophagoides pteronyssinus*, and natural rubber latex (Soluprick SQ 10 HEP, ALK, Copenhagen, Denmark). A reaction with a

Table 1. Subject characteristics by group including lung function, bronchial hyper-responsiveness (BHR), and grade of sputum eosinophilia

	Patients with atopic dermatitis		Control subjects	P-value
<i>N</i>	86	49		
Males, <i>N</i> (%)	29 (34)	36 (74)		< 0.0001 ^x
Age, mean ± SD	25.7 ± 9.3	26.9 ± 6.1		
Smokers, <i>N</i> (%)	17 (20)	5 (10)		0.03 ^x
Body area affected by AD, %, mean ± SD	29 ± 24.4			
Prick test positive, <i>N</i> (%)	72 (84)	18 (37)		< 0.0001 ^x
S-IgE (kU/L), mean ± SD	2498 ± 4157			
Current asthma symptoms, % of subjects	53 (1.7 ± 2.1)	16		< 0.0001 ^x
(mean visual analogue scale/cm ± SD)				
Physician diagnosed asthma, <i>N</i> (%)	31(36)	1(2)		< 0.0001 ^x
Current nasal symptoms, % of subjects	67 (3.1 ± 3.1)	16		< 0.0001 ^x
(mean visual analogue scale/cm ± SD)				
Physician diagnosed allergic rhinitis; <i>N</i> (%)	39 (45)	3 (6)		0.0003 ^x
FEV ₁ (L) mean ± SD	3.80 ± 0.8	4.37 ± 0.86		< 0.0001 ^m
FEV ₁ (%) mean ± SD	95.7 ± 11.6	94.4 ± 12.3		
BHR				
Number tested*	85	49		
Hyper-responsive; <i>N</i> (%)	43 (51)	5 (10)		< 0.0001 ^f
Mild BHR; <i>N</i> (%)	28 (33)	5 (10)		
Moderate BHR; <i>N</i> (%)	10 (12)	0		
Severe BHR; <i>N</i> (%)	5 (6)	0		
Log slope (%) mg	2.32	1.19		< 0.0001 ^t
PD15FEV ₁ (mg)	1.91	2.97		< 0.0001 ^m
Sputum eosinophilia; <i>N</i> (%)	55 (81)	2 (11)		< 0.0001 ^f
Number tested successfully	68	18		
Grade 1; <i>N</i> (%)	38 (56)	2 (11)		
Grade 2; <i>N</i> (%)	10 (15)	0		
Grade 3–4; <i>N</i> (%)	7 (10)	0		

*One patient with physician-diagnosed asthma showed severe obstruction in spirometry and was not further tested for BHR for safety reasons. *t*, *t*-test; *m*, Mann–Whitney *U*-test; *x*, χ^2 test; *f*, Fischer's exact test; AD, atopic dermatitis; s-IgE, serum immunoglobulin E; FEV₁, forced expiratory volume in 1 s; PD15FEV₁, producing a decrease of 15% in FEV₁.

diameter of at least 3 mm in the presence of expected results with control solutions was interpreted as positive for the allergen in question. The subject was regarded as atopic if at least one allergen gave a positive result.

To assess the presence and severity of airway disease, the subjects filled a questionnaire, underwent pulmonary function tests including bronchial challenge with histamine to establish BHR, and underwent an induced sputum test to establish bronchial eosinophilic inflammation.

Current and earlier respiratory symptoms

For all subjects, a previously validated questionnaire was chosen for estimating the prevalence of physician-diagnosed asthma and AR [17]. The degree of asthma symptoms such as cough, wheezing, and dyspnoea, and of nasal symptoms experienced during the preceding year by the patients were estimated using a visual analogue scale from 0 to 10 cm. Patients with symptoms of at least 1 cm on the visual analogue scale were considered to have current asthma or nasal symptoms. The control subjects gave only qualitative answers on the presence of asthma or nasal symptoms.

Lung function tests

The forced expiratory volume in 1 s (FEV₁) [18] was recorded with a flow-volume spirometer (Medikro 905, Medikro Oy, Kuopio, Finland). The results were expressed as percentages (FEV₁%) of national predicted values [19]. BHR was evaluated with a dosimetric histamine challenge test [20] using increasing non-cumulative inhaled doses of 0.025, 0.1, 0.4, and 1.6 mg buffered histamine diphosphate. By using the dose-response curve, the dose of inhaled histamine producing a decrease of 15% in FEV₁ (PD15FEV₁) was determined. According to PD15FEV₁, the BHR was graded as mild (0.41–1.60 mg), moderate (0.11–0.40 mg), or severe (< 0.1 mg) [20]. Subjects with PD15FEV₁ value > 1.6 mg were considered to have normal bronchial responsiveness. Additionally, the result was expressed in terms of the log slope of the dose-response curve. The slope is a continuous and log-normally distributed index [21]. At the time of lung function testing, all subjects had been free from respiratory tract infections for at least the preceding 2 weeks, and short-acting β_2 -agonists had been withheld for at least the previous 12 h.

Sputum induction and analysis

Sputum was induced by inhalation of 5 mL of 3% NaCl solution, using an ultrasonic nebulizer (Omron U1, Omron, Germany) for 15 min as described previously [13]. All patients and 19 control subjects participated. They were pretreated with 200 µg salbutamol (Buventol Easyhaler 100 µg /dose[®] Orion Pharma, Espoo, Finland), by inhalation. Peak expiratory flow (PEF) values were measured before and after induction, to ensure safety of the procedure. Subjects were asked to cough during and after inhalation, and sputum samples were collected. If the PEF fell by more than 15% or if troublesome symptoms appeared, patients were treated with an additional 200 µg of inhaled salbutamol.

The more viscous parts of the sputum samples were collected and mixed. A part of each sample was divided

into two air-dried smears, which were stained with eosin and methylene blue. Cell proportions were assessed semi-quantitatively on a scale from 0 to 4 as described previously [12]. Eosinophilia was graded as relative proportions of eosinophils of all non-squamous cells: approximately, grades 0–4 correspond to eosinophils accounting for < 1%, 1–5%, 6–10%, 11–50%, and > 50%, respectively. Grades 1–4 were considered to represent bronchial eosinophilia. A sample was considered to be adequate and to originate from the lower airways if it contained macrophages and less than 50% of squamous epithelial cells. All analyses were conducted blinded to the clinical characteristics of the subject.

Statistics

Statistical calculations were performed using the SAS Statview program. Groups were compared as required by the distribution of the variable in question using either the *t*-test, the ANOVA, the Mann-Whitney *U*-test, the χ^2 -test, or Fischer's exact test. The relationships of the signs of airway disease with AD were analysed by logistic regression analysis with a fixed model design. The effects were given as odds ratios (OR) with 95% confidence intervals (CI). Spearman's correlation analysis was used to estimate the relationship between variables. A *P*-value less than 0.05 was considered significant.

Results

Airway disease in atopic dermatitis patients and controls

The subject characteristics and results are summarized in Table 1. Patients with AD had more current asthma symptoms, more current nasal symptoms, more physician-diagnosed asthma (36% vs. 2%, OR 10.1, CI 1.3–79.7, *P* = 0.03), and physician-diagnosed AR (45% vs. 6%, OR 4.5, CI 1.2–16.7, *P* = 0.02, fixed model logistic regression) than control subjects. A larger proportion of AD patients (84%) than control subjects (37%) were prick test positive. Nine of the AD patients with physician-diagnosed asthma used inhaled corticosteroids and 18 bronchodilators alone. The only control subject with physician-diagnosed asthma had used inhaled corticosteroids earlier. Six AD patients and one control subject with physician-diagnosed rhinitis used intranasal corticosteroids, and 27 AD patients and two control subjects used oral or topical antihistamines.

Increased BHR was observed more often in AD patients than in control subjects. According to the logistic regression analysis, the risk for physician-diagnosed asthma or AR was significantly increased, and the risk for BHR in AD patients was fivefold compared with the control subjects. Also, the degree of BHR, expressed as mild, moderate, or severe, or as the slope of the dose-response curve, was

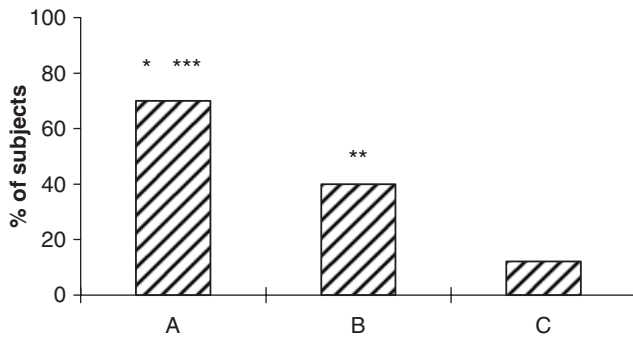


Fig. 1. Bronchial hyper-responsiveness in atopic dermatitis (AD) patients (A) with physician diagnosed asthma; $n=30$, (B) without physician-diagnosed asthma; $n=55$, and (C) control subjects; $n=49$. * $P < 0.0001$ compared with control subjects; ** $P=0.001$ compared with control subjects; *** $P=0.008$ compared with AD patients without physician-diagnosed asthma.

more severe in patients with AD. Of the 30 AD patients with physician-diagnosed asthma, 21 (70%) showed increased BHR. BHR was less prevalent in AD patients without physician-diagnosed asthma than in AD patients with physician-diagnosed asthma ($P=0.008$), but more prevalent than in control subjects ($P < 0.001$, Fischer's exact test) (Fig. 1). Current asthma and nasal symptoms were significantly associated with BHR ($P=0.03$, Spearman's correlation analysis).

An acceptable sputum sample was obtained from 68 (79%) of the AD patients and 18 of the 19 control subjects (95%) volunteering the sputum induction. No significant adverse events or fall in PEF values occurred during the induction. The prevalence of sputum eosinophilia was 81% in AD patients and 11 (%) in control subjects. The risk for sputum eosinophilia was 76-fold compared with the control subjects (fixed model logistic analysis). Differences in sputum eosinophilia were not observed between AD patients with and without physician-diagnosed asthma (Fig. 2). Moderate or severe BHR was more common in patients with sputum eosinophilia than in patients with no sputum eosinophilia ($P=0.0007$, Fischer's exact test). In

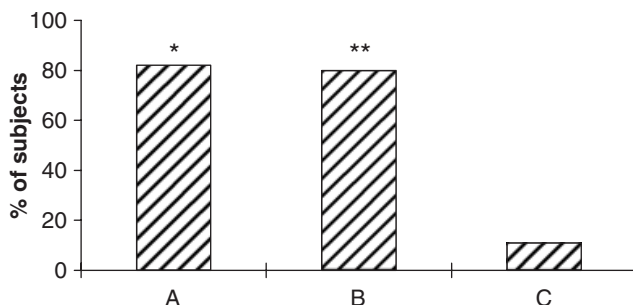


Fig. 2. Sputum eosinophilia in atopic dermatitis patients (A) with physician-diagnosed asthma; $n=19/23$, (B) without physician-diagnosed asthma; $n=36/45$, and (C) control subjects; $n=2/18$ (C). * $P < 0.0001$; ** $P < 0.0001$ compared with control subjects.

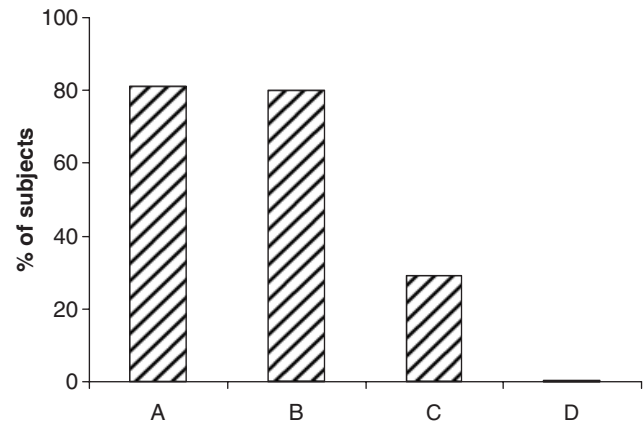


Fig. 3. Eosinophils present in induced sputum in (A) skin prick test (SPT)-positive atopic dermatitis (AD) patients; $n=47/58$, (B) SPT-negative AD patients; $n=8/10$, (C) SPT-positive control subjects; $n=2/7$, and (D) SPT-negative control subjects; $n=0/11$. (A) vs. (D) $P < 0.0001$, (B) vs. (D) $P=0.047$, (A) vs. (C) $P=0.003$, (B) vs. (C) $P=0.18$, NS.

AD patients, sputum eosinophilia was associated with current asthma ($P=0.01$), and nasal ($P=0.02$, Mann-Whitney U -test) symptoms as measured using the visual analogue scale, but they were not significantly associated with physician-diagnosed asthma or physician-diagnosed AR.

The patient and control groups differed slightly in terms of gender and smoking status. It could be shown that gender or smoking did not have an effect on the occurrence or severity of BHR or sputum eosinophilia (ANOVA). After excluding the smoking subjects from both groups, the differences in the prevalences of BHR and sputum eosinophilia remained ($P < 0.0001$, Fischer's exact test).

As 37% of the control subjects had positive prick tests, both the AD and control groups were further divided into SPT positive and negative groups. Patients with AD and positive SPTs had more current asthma symptoms, current nasal symptoms, physician-diagnosed asthma, and physician-diagnosed AR as well as increased BHR and sputum eosinophilia than control subjects with negative SPTs. The differences were more pronounced than between all AD patients and all control subjects (data not shown). Similarly, significant differences were observed between AD patients with positive SPTs and control subjects with positive SPTs as well as between AD patients with negative SPTs and control subjects with negative SPTs (data not shown). Between AD patients with negative SPTs and control subjects with positive SPTs, the significant differences disappeared.

Markers of airway disease in atopic dermatitis patients

SPT-positive AD patients had a higher prevalence of physician-diagnosed asthma ($P=0.01$), physician-diagnosed AR ($P=0.05$, Fischer's exact test), and occurrence of BHR ($P=0.02$, χ^2 -test), but not of current asthma

Table 2. Relationship between airway disease and atopic dermatitis according to logistic regression analysis

Factor	OR	95% CI	P-value
Physician-diagnosed asthma	10.1	1.3–79.7	0.03
Physician-diagnosed allergic rhinitis	4.5	1.2–16.7	0.02
Skin prick test positivity	7.0	2.4–20.7	0.0004
Bronchial hyper-responsiveness	5.5	1.5–20.1	0.01
Sputum eosinophilia	76.1	9.3–623.5	< 0.0001
Reduced FEV ₁	0.5	0.1–2.9	n.s.

OR, odds ratio; CI, confidence intervals.

or nasal symptoms or sputum eosinophils, than SPT-negative AD patients. In SPT-positive AD patients, the mean FEV₁% was lower than in SPT-negative AD patients ($P=0.02$, Mann–Whitney U -test).

s-IgE level was increased in 69% of the AD patients. They had significantly more nasal symptoms ($P=0.04$, Mann–Whitney U -test), physician-diagnosed asthma ($P=0.001$), physician-diagnosed AR ($P=0.0006$), occurrence of BHR ($P=0.02$), and sputum eosinophils ($P=0.03$, Fischer's exact test) than patients with normal s-IgE levels.

Patients with severe AD had a higher mean s-IgE level than patients with moderate AD ($P=0.02$, Mann–Whitney U -test). By using logistic regression analysis, no significant relationship between the severity of AD and respiratory symptoms, physician-diagnosed asthma, or AR, lung function, or sputum eosinophils was found.

Discussion

We found that patients with AD show increased risk of respiratory symptoms, BHR, and eosinophilic airway inflammation. In our series, 51% of the AD patients had BHR and 25% had sputum eosinophilia, suggesting eosinophilic bronchial inflammation. BHR and eosinophilic inflammation may be signs of asthma, conditions that should be identified to enable asthma treatment when needed.

The observed prevalence of BHR in AD patients without physician-diagnosed asthma has varied between 18% and 67% in previous studies. In our series, it was 40%. The variability appears to result from differences in method and age distribution of patients [22–26]. The higher percentages suggest that asthma may often go undiagnosed. Furthermore, AD patients with diagnosed asthma may be undertreated. This appeared to be the case with patients participating in our study. Of the AD patients with physician-diagnosed asthma, 58% used bronchodilating medication as their only treatment, despite 50% of these patients having moderate or severe BHR. This is in disagreement with current guidelines of asthma treatment that outline the importance of anti-inflammatory therapy [27].

An association between airway inflammation and atopy has been demonstrated in previous studies [6–14]. How-

ever, studies on the prevalence of airway inflammation in AD patients have not been reported previously. Estimation of eosinophils in induced sputum is a validated, non-invasive method for the detection of eosinophilic airway inflammation [28]. Such inflammation occurs not only in asthma but also in other airway diseases such as chronic cough, AR, and COPD [11]. A further limitation is that useful sputum samples cannot be obtained from all patients. In this study, the success rate was 79%, which is similar to previous reports [12, 13].

Atopy, asthma, and AD tend to cluster. It is known that AD patients with SPT positivity have increased risk to allergic asthma [29] but it remains uncertain whether the non-allergic or allergic forms of asthma and AD [30, 31] are all truly separate diseases or represent different degrees of severity of the diseases. An analogous clustering of positive laboratory results exists. It has been shown in this and previous studies that patients with asthma frequently have BHR, sputum eosinophilia, positive SPTs, and increased serum IgE levels [4, 12, 13, 32–34]. The same is true of patients with AD as shown: SPT positivity and high levels of IgE were significant risk factors for airway disease in these patients.

The relationship between the severity of AD and airway disease is more vague. The severity of AD was found to associate with s-IgE levels as in previous studies [35, 36] but we found no dependence between eczema severity and BHR or sputum eosinophilia; the result is analogous to that of Brinkman *et al.* for BHR [37].

We observed BHR and sputum eosinophilia to occur also in AD patients with negative SPTs or normal s-IgE levels or both. The prevalences were lower than in SPT-positive patients, but higher than in control subjects. Patients with prick test positive AD had clearly higher prevalences of airway disease and related positive test results.

We conclude that AD patients, especially those with positive SPTs and high levels of s-IgE, often also have BHR and an eosinophilic airway inflammation typical of asthma. They frequently have respiratory symptoms, and may be at risk of developing clinical asthma. Longitudinal data are needed to assess the outcome of AD patients with signs of airway disease, in order to identify those who need early initiation of asthma treatment.

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