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Original Article

Clinical symptoms, inflammatory factors and responses to hormones in bronchial asthma patients with different inflammatory phenotypes

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Abstract: Objective: To investigate the differences in clinical symptoms, inflammatory factors and responses to hormones among bronchial asthma patients with different inflammatory phenotypes. Methods: A total of 287 asthmatic patients were enrolled and divided into eosinophilic group (Eos group, n=123), neutrophilic group (Neu group, n=80), mixed granulocytic group (Mix group, n=54) and pauci-granulocytic group (Pau group, n=30) based on the cytological examination of sputum. The general information from the study subjects was collected, and the fractional exhaled nitric oxide (FENO) level and the inflammatory factor level in peripheral blood were determined. Moreover, patients were scored via the asthma control test (ACT), followed by pulmonary function test. Results: The proportion of asthma inflammatory phenotypes in each group was: Eos group (42.86%), Neu group (27.87%), Mix group (18.82%) and Pau group (10.45%). The Neu group exhibited a significantly higher proportion of family history of asthma than the Eos group and Pau group ($P<0.01$), and it was significantly higher in Mix group than that in Pau group ($P<0.05$). The level of FENO in the Eos group was significantly higher than those in the remaining three groups ($P<0.05$). The Eos group had significantly higher levels of interleukin (IL)-5 and IL-13 than the remaining three groups ($P<0.05$), and the level of IL-18 in Neu group was higher than those in the remaining three groups ($P<0.05$). The Pau group exhibited a higher ACT score, peak expiratory flow (PEF)%, and forced expiratory volume in 1 second (FEV1)/forced vital capacity (FVC) ratio (%) than both the Eos group and Neu group ($P<0.05$). The consumption of systemic and inhaled hormones in the Eos group was lower than those in Mix and Pau groups ($P<0.05$). Conclusion: The clinical symptoms, inflammation and response to hormone treatments vary among asthma patients with different inflammatory phenotypes.

Keywords: Asthma, inflammatory phenotypes, inflammatory factors, hormones

Introduction

Bronchial asthma (asthma) is a common chronic disorder of the airways that is complex and characterized by variable and recurring symptoms, airflow obstruction, bronchial hyperresponsiveness, and an underlying inflammation [1, 2]. The pathogenesis of asthma is very complicated involving several factors. Airflow limitation in asthma is recurrent and caused by a variety of changes in the airway including bronchoconstriction, airway edema, airway hyperresponsiveness, airway remodeling [3]. Previous studies have suggested that this airway inflammation is mainly induced by the filtration of eosinophils [4-6], while recent studies have discovered that numerous neutrophils infiltrate the sputum of some patients, indicating that there are different airway inflammatory phenotypes in asthma [7-10]. Asthma can be classified into four kinds of inflammatory phenotypes according to the classification of induced sputum cells [11, 12]. Currently, there are a few studies on the clinical symptoms, the level of inflammation and the clinical effects of hormone treatments among asthmatic patients with different inflammatory phenotypes. Therefore, the present study aims to explore these, hoping to provide more reasonable and effective clinical therapeutic guidance for asth-

Table 1. Distribution frequency of each group

Group	n (%)
Eos group	123 (42.86)
Neu group	80 (27.87)
Mix group	54 (18.82)
Pau group	30 (10.45)

ma in patients with different inflammatory phenotypes.

Information and methods

Study subjects

Asthmatic patients admitted to the Respiratory Medicine Department of The Sixth People's Hospital of Nantong from January 2016 to 2018 were selected, and the included patients met the following criteria: 1) patients conformed to the diagnostic criteria for bronchial asthma formulated by the Chinese Medical Association, 2) those without history of foreign body aspiration and 3) those with favorable compliance and complete information. Exclusion criteria: 1) patients complicated with other wheezing diseases in the respiratory tract, 2) those complicated with infectious diseases in other body systems or 3) those with the dysfunction of the heart, kidney, liver or other major organs.

A total of 287 asthmatic patients were included according to the above criteria. With the percentage of eosnophils in induced sputum (3%) and that of neutrophils (63%) as the cut-off values, the eligible patients were divided into four groups: eosinophilic group [Eos group, no less than 3% eosinophils and less than 63% neutrophils, including 123 patients aged (41.3±11.2) years old on average], neutrophilic group [Neu group, less than 3% eosinophils and no less than 63% neutrophils, including 80 patients aged (40.7±11.4) years old on average], mixed granulocytic group [Mix group, no less than 3% eosinophils and less than 63% neutrophils, including 54 patients aged (45.3±11.7) years old on average] and pauci-granulocytic group [Pau group, less than 3% eosinophils and 63% neutrophils, including 30 patients aged (43.5±12.8) years old on average]; all based on the cytological examination results of induced sputum. All the subjects signed the informed consent. This study was approved by the ethnic committee of our hospital.

Study methods

Collection of information: The name, sex, age, symptoms, signs, course of disease, body mass index (BMI), family history, atopy, smoking history and response to hormone treatments of the subjects were collected. The fractional exhaled nitric oxide (FENO) level was determined with the FENO tester, and 3 mL venous blood was collected from the elbow of the patients to measure the levels of interleukin (IL)-5, IL-8, IL-13 and IL-17 in peripheral blood using a commercial kit according to manufacturer's instructions, and wea also conducted an asthma control test (ACT) score. Afterwards, the forced expiratory volume in 1 second (FEV1)/the predicated value ratio (FEV1%), the daily variability of peak expiratory flow (PEF%) and the FEV1/forced vital capacity (FVC) ratio (FEV1/FVC)% before and after hormone treatments were measured using the pulmonary function analyzer. The attending physicians from the Respiratory Department carried out the above operations.

Statistical methods

SPSS 20.0 software was used for statistical analysis. Chi-square test was adopted for the comparison of count data between two groups. The results of measurement data were expressed as mean ± standard deviation (SD). Non-paired *t* test and one-way analysis of variance were conducted for the comparisons of measurement data between two groups and among groups, respectively. $P<0.05$ represented a difference that was statistically significant.

Results

Distributions of asthma inflammatory phenotypes in each group

The proportion of asthma inflammatory phenotypes in the Eos group (n=123), Neu group (n=80), Mix group (n=54) and Pau group (n=30) were 42.86%, 27.87%, 18.82% and 10.45%, respectively (**Table 1**).

Comparison of general information

The Neu group had a significantly higher proportion of family history of asthma than the Eos

Table 2. Comparison of general information among all groups

Group	n	Female/ male (n)	Age (year old)	Course of disease (year)	BMI (kg/m^2)	Family history [n (%)]	Atopy [n (%)]	Smoking history [n (%)]
Eos group	123	74/43	41.3±11.2	7.5 (2.0, 21.0)	23.4±3.4	27 (22.0)	68 (55.3)	19 (15.5)
Neu group	80	47/33	40.7±11.4	5.0 (1.0, 16.0)	23.5±3.5	34 (42.5)^{aa}	36 (45.0)	9 (11.3)
Mix group	54	31/23	45.3±11.7	8.0 (2.0, 19.0)	23.8±4.0	18 (33.33)	26 (48.2)	14 (25.9)
Pau group	30	23/7	43.5±12.8	9.5 (5.0, 18.5)	23.7±3.1	4 (13.3)^{bb,c}	18 (60.0)	7 (23.3)

Note: $^{aa}P<0.05$, vs. Eos group, $^{bb}P<0.01$, vs. Neu group and $^{c}P<0.05$, vs. Mix group.

Table 3. Comparisons of FENO levels among all groups

Group	Eos group	Neu group	Mix group	Pau group
n	123	80	54	30
FENO level (ppb)	83.0±59.2	25.4±19.4^{a}	21.7±18.6^{a}	36.3±19.8^{a}

Note: $^{a}P<0.05$, vs. Eos group.

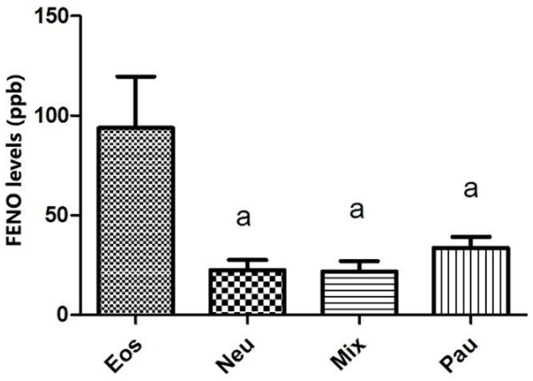

Figure 1. Comparisons of FENO levels among all groups ($^{a}P<0.05$, vs. Eos group).

and Pau groups ($P<0.05$), and there were no significant differences in the other indicators (**Table 2**).

Comparison of FENO level

The Eos group had a significantly higher level of FENO than the remaining three groups ($P<0.05$) (**Table 3**; **Figure 1**).

Comparison of inflammatory factor levels

The levels of IL-5 and IL-13 in the Eos group were significantly higher than those in the remaining three groups ($P<0.05$), so was the level of IL-8 in the Neu group ($P<0.05$). However, there was no difference of the level of IL-17 among all groups ($P>0.05$) (**Table 4**).

Comparison of ACT score

The Pau group had a significantly higher ACT score than the Eos and Neu groups ($P<0.05$) (**Table 5**; **Figure 2**).

Comparisons of pulmonary functions

The Pau group exhibited a significantly higher PEF% and FEV1/FVC ratio (%) than the Eos and Neu groups ($P<0.05$) (**Table 6**).

Use of hormones

The consumption of systemic and inhaled hormones in the Eos group was significantly lower than those in the Mix and Pau groups ($P<0.05$). There was no difference in the duration of usage of systemic and inhaled hormones among all groups ($P>0.05$) (**Table 7**).

Discussion

Asthma is characterized by different clinical phenotypes through pathological and cytological detection, with both the infiltration of eosnophils and substantial neutrophilic granulocytosis in the airway of asthmatic patients [13-17]. In this study, the most common inflammatory phenotype was the Eos type, accounting for 42.86%, followed by the Neu type, Mix type and Pau type. The Neu group and Mix group exhibited a high proportion of family history of asthma, indicating that the asthmatic patients with family history of the disease are much more likely to experience neutrophilic granulocytosis in their sputum.

The main pathogenesis of asthma is airway inflammation, and the airway epithelium is the main source of FENO, which has become one of the airway inflammation markers for evaluating the degree of inflammation in asthma [18-20]. The results of this study showed that the level of FENO in the Eos group was significantly higher than those in the remaining three groups; suggesting that FENO is involved in the airway inflammatory responses, and the increase in its level is more closely related to the severity of Eos-type asthma. In the clinical diagnosis of

Table 4. Comparisons of inflammatory factor levels among all groups

Group	n	IL-5 (ng/L)	IL-8 (ng/L)	IL-13 (ng/L)	IL-17 (ng/L)
Eos group	123	28.2±14.6	92.8±64.8	41.8±14.8	16.0±7.4
Neu group	80	18.4±6.6[a]	135.6±80.8[a]	33.8±7.4[a]	17.4±10.3
Mix group	54	18.6±7.8[a]	92.6±63.3[b]	36.3±7.8[a]	16.4±5.9
Pau group	30	20.2±9.8[a]	98.6±52.5[b]	35.2±8.0[a]	16.2±6.7

Note: [a]P<0.05, vs. Eos group and [b]P<0.05, vs. Neu group.

Table 5. Comparisons of ACT scores among all groups

Group	Eos group	Neu group	Mix group	Pau group
n	123	80	54	30
ACT score	16.3±3.0	17.2±3.4	19.0±3.2	19.2±3.0[a,b]

Note: [a]P<0.05, vs. Eos group and [b]P<0.05, vs. Neu group.

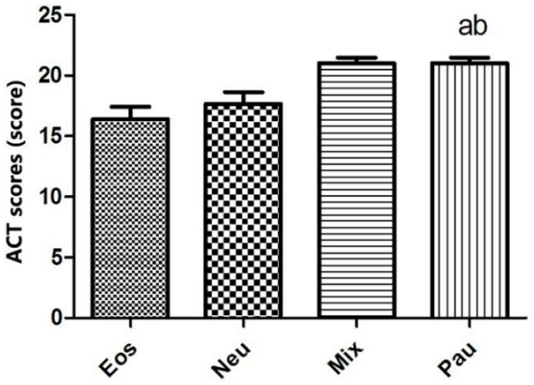

Figure 2. Comparisons of ACT scores among all groups ([a]P<0.05, vs. Eos group and [b]P<0.05, vs. Neu group).

asthma, the severity of different airway inflammatory phenotypes can be assessed through determining the level of FENO.

The levels of IL-5 and IL-13 in the Eos group were significantly higher than those in the remaining three groups, so was the level of IL-8 in the Neu group. IL-5 and IL-13 are important inflammatory mediators in the T helper (Th) 2 immune pathway. The former can stimulate the proliferation, differentiation and activation of eosnophils, while the latter can promote the migration of eosinophils from blood to the airway by facilitating the secretion of their chemotactic factors [21]. The elevated IL-5 and IL-3 levels in this study indicated that Eos-type asthma is associated with the Th2 immune pathway. IL-8, as a key inflammatory factor in the Th17 immune pathway, it can attract and activate neutrophils, and it releases active products to control inflammation after binding to neutrophils. The level of IL-8 in the Neu group was significantly higher than those in the remaining three groups, indicating that Neu-type asthma is correlated with the Th17 immune pathway.

According to literature reports in recent years, the release of eosinophils and neutrophils is associated with the damage, detachment and highly-secreted mucus from airway epithelial cells, the former two of which can be caused by the infiltration of eosnophils, and Eos-type asthma exhibits more obvious airway hyperresponsiveness and subepithelial fibrosis than non-Eos type asthma [22]. The inflammatory mediators released by neutrophils in sputum are positively correlated with airway epithelial damage and excessive mucus secretion. In this study, both the Eos group and Neu group had lower ACT scores and worse pulmonary function than the Pau group, and the possible cause might be that the Eos group and Neu group released more eosnophils and neutrophils, respectively; leading to more marked damage and detachment of airway epithelial cells and more sever airway inflammatory responses and airflow obstruction. Hence, it was more difficult to control asthma and pulmonary functions were worse. Currently, there is still a scarcity of systematic and comprehensive studies on the asthma responses with different inflammatory phenotypes to hormone treatments. Bacci et al. [23] found that compared with non-Eos-type asthma, the Eos-type exhibits a better response to inhaled hormone treatments, while the study of Jiang Yuwei et al. revealed that [24] glucocorticoid treatment is remarkably more effective for the asthmatic patients with a higher proportion of eosnophils in the sputum.

Conclusion

The Eos group had lower consumption of systematic and inhaled hormones than the Mix group and Pau group, suggesting that glucocorticoid treatment manifests a more favorable efficacy in Eos-type asthma than that in Mix-type and Pau-type asthma. The patients in the Neu, Pau and Mix groups, especially those in

Table 6. Comparisons of pulmonary functions among all groups

Group	n	FEV1%	PEF%	FEV1/FVC ratio (%)
Eos group	123	64.1±25.8	71.8±30.9	59.7±17.4
Neu group	80	73.2±24.7	61.5±25.2	61.8±15.8
Mix group	54	82.3±24.3	80.8±23.8	64.5±12.7
Pau group	30	83.1±24.0	83.5±24.1[a,b]	69.4±13.6[a,b]

Note: [a]*P*<0.05, vs. Eos group and [b]*P*<0.05, vs. Neu group.

Table 7. Comparison of hormone use

Group	n	Consumption of systemic hormone (mg)	Use time of systemic hormone (d)	Consumption of inhaled hormone (mg)	Use time of inhaled hormone (d)
Eos group	123	255.2±201.5	5.6±3.4	25.5±28.4	7.6±4.3
Neu group	80	371.5±248.5	3.8±3.0	29.4±20.8	6.2±4.6
Mix group	54	621.7±430.9[a]	6.7±4.7	53.2±38.6[a]	9.8±4.8
Pau group	30	617.3±428.8[a]	6.2±3.0	33.8±30.5[a]	7.5±4.0

Note: [a]*P*<0.05, vs. Eos group.

the latter two groups, needed to take higher-dose hormones to alleviate asthma. Therefore, in the clinical treatment, patients with Eos-type asthma should be administrated with low-dose glucocorticoids, while those with Pau-type and Mix-type asthma should take high-dose glucocorticoids.

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Disclosure of conflict of interest

None.

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