Serum level of eosinophil-derived neurotoxin: a biomarker for asthma severity in adult asthmatics

by

Youngsoo Lee

Major in Medicine

Department of Medical Sciences

The Graduate School, Ajou University

Serum level of eosinophil-derived neurotoxin:

a biomarker for asthma severity in adult asthmatics

by

Youngsoo Lee

A Dissertation Submitted to The Graduate School of Ajou University in Partial Fulfillment of the Requirements for the Degree of Master of Medicine

Supervised by

Hae-Sim Park, M.D., Ph.D.

Major in Medicine

Department of Medical Sciences

The Graduate School, Ajou University

February 2019

This certifies that the dissertation of Youngsoo Lee is approved.

SUPERVISORY COMMITTEE

_____

Hae-Sim Park

_____

Dong-Ho Nahm

_____

Yoo-Seob Shin

The Graduate School, Ajou University

February 2019

- ABSTRACT –

Serum level of eosinophil-derived neurotoxin: a biomarker for asthma severity in adult asthmatics

Background: Severe asthma is a refractory disease with complex and heterogeneous pathophysiologic mechanisms. Eosinophilic inflammation is considered a key component of severe asthma (SA). However, there has been no reliable biomarker representing the eosinophilic inflammation of SA.

Objective: This study aimed to characterize the clinical features of SA and to evaluate whether serum eosinophil-derived neurotoxin (EDN) level may predict the eosinophilic inflammation of SA in adult asthmatics.

Methods: Severe asthmatics (n = 235) and nonsevere asthmatics (n = 898) were enrolled from Ajou University Hospital, South Korea. Serum levels of EDN and periostin (type 2 inflammatory biomarkers) were measured using ELISA, and their association with clinical parameters was analyzed.

Results: Severe asthmatics were older and had longer disease durations with significantly lower levels of FEV_1% pred and methacholine PC_{20}. Total eosinophil count (TEC) and sputum eosinophil count (%) were significantly higher in severe asthmatics than in nonsevere asthmatics. Serum EDN level and serum periostin level were significantly higher in severe asthmatics than in nonsevere asthmatics ($P < 0.034$ and $P < 0.001$, respectively). Correlations of serum EDN level to TEC ($r = 0.319$, $P < 0.001$) and to serum periostin level ($r = 0.302$, $P < 0.001$) were noted in total asthmatics.

Conclusion: These findings suggest that serum EDN level can be a useful biomarker for predicting the phenotype of SA in adult asthmatics.

Keywords: EDN; severe asthma; eosinophilic inflammation; biomarker

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

I. INTRODUCTION

Asthma is a chronic inflammatory disease of multifactorial etiologies that affects 300 million people worldwide. Initially, asthma was considered typical eosinophilic airway inflammation initiated by allergic sensitization, which results in airway hyperresponsiveness (AHR) and acute bronchoconstriction. Among asthma patients, severe asthma (SA) is responsible for more than 50% of the medical expense of asthma, even though it accounts for 5%-10% of entire asthma patients. Severe asthmatics are suffering from frequent exacerbations that contribute to progressive lung function decline and increasing burden of medical cost (1).

The eosinophilic phenotype of asthma is related to type 2 cytokines such as IL-4, L-3, and IL-13. Corticosteroid (CS) is a key anti-inflammatory agent to control eosinophilic inflammation, and biologics targeting type 2 inflammation are effective in SA with high total eosinophil count (TEC) (2, 3). There have been traditional biomarkers used to monitor eosinophilic inflammation in asthma including TEC, sputum eosinophil count (%), serum periostin level and fractional excretion of nitric oxide (FeNO) concentration (4). Not only are sputum eosinophil count and FeNO concentration relatively hard to yield and variable by CS therapy (5-7), but also the studies of TEC predicting sputum eosinophil counts in eosinophilic asthma have shown conflicting results (8-10). Since there has been no biomarker available that can effectively reflect chronic eosinophilic inflammation in SA, the need for identifying an appropriate biomarker is continuously emerging. While periostin has been known to be related to eosinophilic inflammation in asthma, several studies have reported eosinophil-derived neurotoxin (EDN), a degranulation protein released from eosinophils, is deemed to represent eosinophil activation in childhood asthma and atopic dermatitis, and its value as a serum biomarker has not yet been evaluated in adult severe asthmatics (11, 12).

We hypothesized that serum EDN might predict the phenotype of SA and analyzed its association with eosinophil-related clinical parameters in patients with SA compared to those with nonsevere (NSA, mild to moderate) asthma.

II. MATERIALS AND METHODS

A. STUDY SUBJECTS

A total of 1,133 Korean adult asthmatic patients, including 898 of severe and 235 of nonsevere asthmatics, were enrolled in the study. Asthma was diagnosed according to the Global Initiative for Asthma (GINA) guideline based on asthma symptoms and lung function test results. The subjects were divided into the SA and NSA groups by the International European Respiratory Society (ERS)/American Thoracic Society (ATS) guidelines (13). Any patient who had underlying respiratory diseases other than asthma was excluded. All participants provided written informed consent before participating in this study, and Institutional Review Board approval was obtained (AJIRB-GEN-SMP-13).

Atopy was defined as at least 1 positive reaction to a skin prick test of 55 common inhalant allergens (Bencard Co., Brentford, UK), compared to histamine and saline controls. Pulmonary function test was done as previously described (14). A methacholine bronchial challenge test was performed using doubled doses of Provocholine® (methacholine chloride USP, Methapharm Inc., FL, USA) from 1 to 16 mg/mL. Serum total IgE and specific IgE was measured by ImmunoCAP® system (ThermoFisher Scientific, Waltham, MA, USA). Chronic rhinosinusitis and nasal polyps were diagnosed by paranasal sinus radiography, computed tomography, and rhinoscopic findings. Sputum eosinophil and neutrophil counts (%) were expressed as the percentage of the cells among non-squamous cells in sputum.

B. MEASUREMENT OF EOSINOPHILIC BIOMARKERS

Serum samples from the patients were collected on the initial evaluation and stored at −80ºC. Serum EDN level (MBL International, Woburn, MA, USA) and serum periostin level (Shino-Test, Kanagawa, Japan) were measured by enzyme-linked immunosorbent assay (ELISA) according to the manufacturer's protocols.

C. STATISTICAL ANALYSIS

Student's *t*-test was used to compare continuous variables, and Pearson's chi-square test was done for comparing categorical variables. Statistical correlation was analyzed by Pearson's correlation analysis. All statistical analyses were done by using IBM SPSS software version 18.0 (IBM Corp., Armonk, NY, USA). *P* values < 0.05 were considered statistically significant.

III. RESULTS

A. DEMOGRAPHIC AND CLINICAL CHARACTERISTICS

Demographic and clinical characteristics of severe asthmatics (n=235) and nonsevere asthmatics (n=898) were analyzed (Table 1). Severe asthmatics were older (46.01 ± 13.93 *vs*. 42.69 ± 14.63 years, $P = 0.001$) and had a longer asthma duration (8.04 ± 6.61 *vs*. 5.72 ± 10.56 years, $P < 0.001$) than nonsevere asthmatics. They had higher prevalence of upper airway comorbidities such as paranasal sinusitis (50.0% *vs*. 35.0%, $P = 0.008$) and nasal polyps (41.7% *vs*. 28.0%, $P = 0.007$) than nonsevere asthmatics. However, no differences were noted in proportions of female sex, atopy or smoking history between severe and nonsevere asthmatics (62.55% *vs*. 61.87%, $P = 0.848$; 53.33% *vs*. 54.48%, $P = 0.885$; 48.4% *vs*. 40.1%, $P = 0.095$; respectively). Severe asthmatics had lower levels of baseline FEV_1% pred. (71.38% ± 21.81 % *vs*. 89.75% ± 20.29 %, $P < 0.001$) and methacholine PC_{20} (7.37 ± 12.15 *vs*. 10.79 ± 20.09 mg/mL, $P = 0.042$) than nonsevere asthmatics. TEC (542.01 ± 832.51/μL *vs*. 407.97 ± 754.47/μL, $P = 0.002$) and sputum eosinophil (26.95% ± 35.59% *vs*. 22.11 ± 33.15%, $P = 0.031$) were higher in severe asthmatics than in nonsevere asthmatics, whereas serum total IgE level (368.01 ± 534.32 *vs*. 389.17 ± 690.61 kU/L, $P = 0.210$) and neutrophil counts (56.62% ± 35.29% *vs*. 58.90% ± 33.84%, $P = 0.558$) were not significantly different.

Table 1. Clinical characteristics of severe and nonsevere asthmatics

	Severe asthmatics	Nonsevere asthmatics	*P* value
Age (yr-old)	46.01 ± 13.93	42.69 ± 14.63	0.001
Female sex	62.55%	61.87%	0.848
Atopy	53.33%	54.48%	0.885
Current or ex-smoker	48.4%	40.1%	0.095
Asthma duration (yr)	8.04 ± 6.61	5.72 ± 10.56	< 0.001
Paranasal sinusitis	50.0%	35.0%	0.008
Nasal polyp	41.7%	28.0%	0.007
Baseline FEV_1 (%)	71.38 ± 21.81	89.75 ± 20.29	< 0.001
methacholine PC_{20} (mg/mL)	7.37 ± 12.15	10.79 ± 20.09	0.042
Total IgE (kU/L)	368.01 ± 534.32	389.17 ± 690.61	0.210
Total eosinophil count (/μL)	542.01 ± 832.51	407.97 ± 754.47	0.002
Sputum eosinophil (%)	26.95 ± 35.59	22.11 ± 33.15	0.031
Sputum neutrophil (%)	56.62 ± 35.29	58.90 ± 33.84	0.558
Serum EDN level (ng/mL)	69.08 ± 42.40	58.46 ± 35.56	0.034
Serum periostin level	91.56 ± 41.21	77.72 ± 38.56	0.001

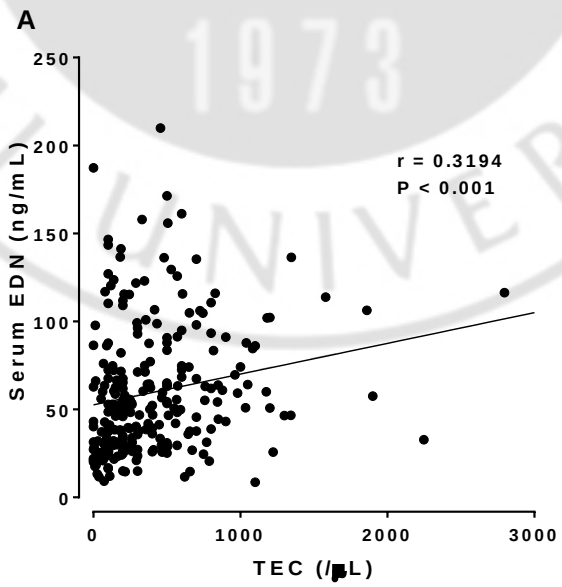
Each value is presented as mean ± standard deviation or % counted.

P value was calculated by Student's *t*-test or the Pearson's chi-square test.

FEV_1, forced expiratory volume in 1 second; FVC, forced vital capacity; PC_{20}, provocation concentration causing a 20% fall in FEV_1; pred, predicted; EDN, eosinophil-derived neurotoxin.

B. ASSOCIATIONS BETWEEN CLINICAL PARAMETERS

The serum levels of EDN and periostin were significantly higher in patients with SA than in those with NSA (69.08 ± 42.40 *vs*. 58.46 ± 35.56 ng/mL, $P = 0.034$; 91.56 ± 41.21 *vs*. 77.72 ± 38.56 ng/mL, $P = 0.001$) (Table 1). In total asthmatics, serum EDN level showed a significant correlation with TEC ($r = 0.319$, $P < 0.001$; Fig. 1A), while serum periostin level was less correlated with TEC ($r = 0.222$, $P < 0.001$; Fig. 1B). Moreover, the serum EDN level had a positive correlation with the serum periostin level in total asthmatics ($r = 0.302$, $P < 0.001$, Fig.1C). When asthmatics were classified according to serum EDN levels: the high responders to serum EDN (having higher levels than mean + 1 SD of serum EDN of the study subjects) showed a higher TEC (971.19 ± 1550.92/μL *vs*. 367.47 ± 347.01/μL, $P = 0.024$) and a lower baseline FEV_1% pred (84.29% ± 20.80% *vs*. 90.46 ± 19.05%, $P = 0.057$) than low responders (having serum EDN levels less than mean + 1 SD). High responders to serum periostin (serum periostin level ≥ mean + 1 SD) showed lower methacholine PC_{20} levels (6.24 ± 8.47 *vs*. 9.92 ± 12.26 mg/mL, $P = 0.008$) and a higher prevalence of SA (35.7% *vs*. 19.5%, $P = 0.002$) than low responders (serum periostin level < mean +1 SD). In addition, the serum EDN level presented the highest area-under-curve (AUC) value as a single biomarker for differentiating eosinophilic (TEC ≥ 300/μL) and noneosinophilic (TEC < 300/μL) asthmatics (AUC = 0.657, $P < 0.001$).



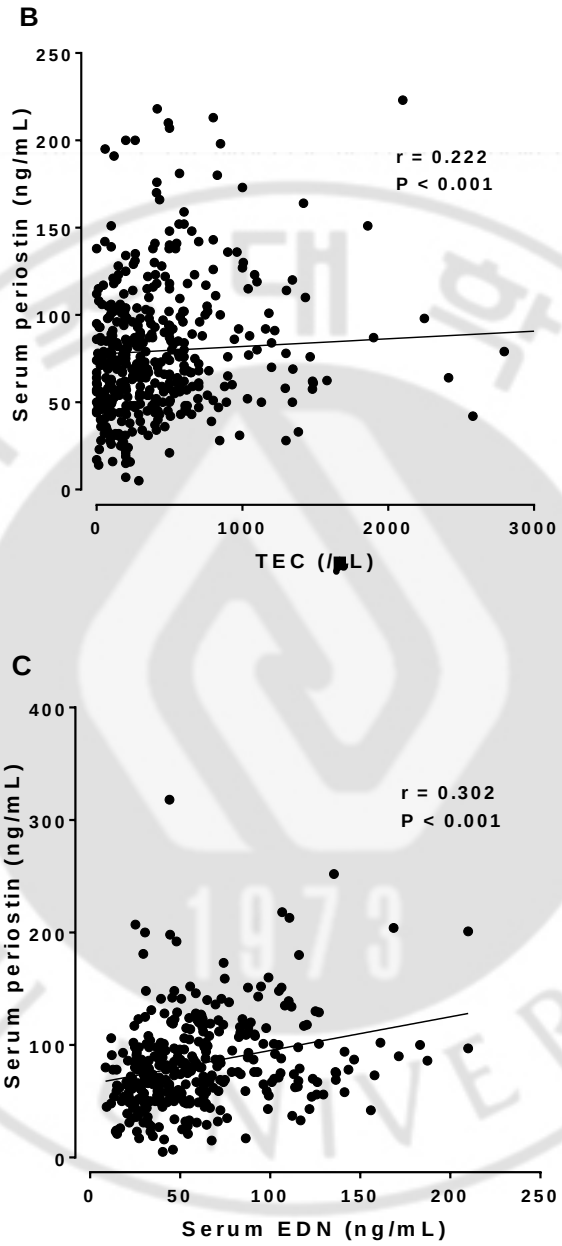


Fig 1. Correlation of the eosinophilic biomarkers in total asthmatics; TEC to serum EDN level (A), TEC to serum periostin level (B), and serum EDN level to serum periostin level (C).

IV. DISCUSSION

Eosinophils are key driving cells in airway inflammation of SA as 50 % of severe asthmatics are associated with persistent elevation of type 2 inflammatory markers. However, the ideal method for assessing type 2 inflammation of SA is not well established.(15, 16) Increased expression of periostin in serum and tissue has been suggested as a biomarker for type 2 inflammation in adult asthmatics.(17) Our previous study demonstrated that the adult asthmatics with eosinophilic asthma or SA had significantly higher serum periostin levels compared to the control groups, indicating that serum periostin is a biomarker for type 2 inflammation in asthmatics.(4) Serum periostin levels were significantly higher in the SA group than in the NSA group, but less correlated to TEC than serum EDN levels. These findings suggest that the serum EDN level could be a more useful biomarker (than previous serum biomarkers suggested) for predicting the eosinophilic phenotype of SA in adult asthmatics.

Recent large-scale studies have extended understanding of the characteristics of SA. They proved distinct demographic and clinical characteristics including older age, female predominance, smoking history, frequent exacerbations, lower baseline lung function, and more need for high doses of inhaled or systemic CS. Also higher prevalence of upper and lower respiratory comorbidities were consistently relevant to SA.(18-20) In this study, we confirmed that severe asthmatics were older and had a longer duration of asthma and higher prevalence of comorbid conditions (paranasal sinusitis and nasal polyps), while no differences were noted in atopy prevalence or smoking history. It also showed lower FEV_1% pred. levels and increased airway hyperresponsiveness as a key feature of SA. Persistent eosinophilic inflammation is another key feature of SA and is closely associated with frequent asthma exacerbations. TEC or sputum eosinophilia (>3%) has been used to define the phenotype of eosinophilic asthma. There was a study in which adult asthmatics with more than 300 /μL of TEC had the highest tendency of asthma exacerbation. (21) Still, another study demonstrated that TEC was rarely correlated with eosinophilia in bronchoalveolar lavage (BAL) fluid or endobronchial biopsies in patients with SA.(22) It is not practical to differentiate the phenotypes using eosinophil-related clinical parameters

as they have been demonstrated to vary widely depending on clinical severity and medications.(20, 23, 24) In this study, TEC and sputum eosinophil count (%) were significantly higher in SA than in NSA. Considering severe asthmatics have poor lung function with severe airway hyperresponsiveness, close monitoring and active therapeutic intervention by applying serum biomarkers are urgent to prevent asthma exacerbations and lung function decline.

It is essential to distinguish the phenotypes between eosinophilic and noneosinophilic asthmatics because more severe eosinophilic inflammation implies the higher risk of SA. Effective control of eosinophilic inflammation was proved beneficial in decreased asthma exacerbations.(25-27) Although sputum eosinophil counts have determined the phenotype of eosinophilic asthma, sputum induction has a risk and is not always successful because sputum differential cell counts are variable.(28) For this reason, serum biomarker that is stable and reproducible to represent type 2 inflammation is considered useful regarding its safety and easy accessibility. Serum EDN level was the only marker for differentiating childhood asthmatic patients with acute exacerbation from those with stable status, showing a greater correlation with the severity of asthma when compared to serum ECP levels or TEC.(29) In another study, it was correlated with the degree of persistent airflow limitation in allergic asthmatics.(30) In this study, serum EDN levels were significantly higher in severe asthmatics than in nonsevere asthmatics; significant positive correlations of serum EDN level to TEC and to serum periostin levels were found in total asthmatics. Moreover, high EDN-responders had lower FEV_1% pred., which suggests that serum EDN level may be a useful biomarker for representing SA. Further investigations are required to explore the exact function of EDN in the pathogenic mechanisms of SA. Moreover, there are other inflammatory mechanisms than eosinophilic inflammation involved in SA. Airway neutrophilia is documented in patients with severe exacerbations, but its role in SA remains to be elucidated.(31) A few studies demonstrated increased IL-17 levels in samples (induced sputum, BAL samples, and bronchial biopsies) of severe asthmatics.(32, 33) There has been an investigation where IL-17-producing cells and IL-17-related gene expression signature were increased and orthogonal to eosinophilic inflammation in SA.(34) Some patients with SA had mixed cellular profiles (both sputum eosinophilia and

neutrophilia) in their sputum. Our previous report demonstrated increased expression of neutrophil extracellular traps (NETs) in severe asthmatics than non-severe asthmatics. NETs were found to induce eosinophil activation suggesting a close interaction between these 2 inflammatory cells.(35) Taken together, we propose that increased serum EDN level may potentially predict 2 phenotypes of SA: eosinophilic inflammation and poor lung function in adult asthmatics, although long-term follow-up studies are essential to monitor changes in serum EDN levels according to the status of airway inflammation and with trials of anti-inflammatory treatment.

Periostin is a matricellular protein that is deposited in the airway subepithelium of asthmatics. Recently, periostin has been emphasized as a biomarker for type 2 airway inflammation and the responses to type 2-targeted therapy in asthmatics.(36) Several studies already suggested that periostin stimulated by IL-13 facilitates the TGF-β signaling pathway, thereby leading to airway fibrosis.(37, 38) Moreover, it has been known that activated eosinophils induce fibroblasts to secrete periostin and to promote fibrosis.(39) Serum periostin levels significantly correlate with the degree of AHR in pediatric asthma.(40) In the present study, serum periostin levels were significantly higher in patients with SA than in those with NSA, and significant associations were found with a degree of AHR and prevalence of SA. Further studies will help determine how EDN and periostin work differently or interact in the pathogenesis of SA.

The limitation of this study is that it was a cross-sectional study in a single tertiary center. Thereby, it was unable to control the medications such as inhaled or systemic CS that affect the eosinophilic inflammation in all the study subjects. Follow-up studies are essential to evaluate the clinical significance of serum EDN responses to anti-inflammatory medications and long-term outcomes.

V. CONCLUSION

These findings suggest that serum EDN level can help identify SA with eosinophilia in adult asthmatics. Considering SA has heterogeneous phenotypes, further studies are needed

to identify useful biomarkers for representing subtypes of SA (eosinophilic, noneosinophlic or mixed type).

REFERENCES

1. Yoo KH, Ahn HR, Park JK, Kim JW, Nam GH, Hong SK, et al. Burden of Respiratory Disease in Korea: An Observational Study on Allergic Rhinitis, Asthma, COPD, and Rhinosinusitis. Allergy, asthma & immunology research. 2016;8(6):527-34.
2. Castro M, Zangrilli J, Wechsler ME, Bateman ED, Brusselle GG, Bardin P, et al. Reslizumab for inadequately controlled asthma with elevated blood eosinophil counts: results from two multicentre, parallel, double-blind, randomised, placebo-controlled, phase 3 trials. The Lancet Respiratory medicine. 2015;3(5):355-66.
3. Pavord ID, Korn S, Howarth P, Bleecker ER, Buhl R, Keene ON, et al. Mepolizumab for severe eosinophilic asthma (DREAM): a multicentre, double-blind, placebo-controlled trial. Lancet (London, England). 2012;380(9842):651-9.
4. Kim MA, Izuhara K, Ohta S, Ono J, Yoon MK, Ban GY, et al. Association of serum periostin with aspirin-exacerbated respiratory disease. Annals of allergy, asthma & immunology : official publication of the American College of Allergy, Asthma, & Immunology. 2014;113(3):314-20.
5. Pin I, Freitag AP, O'Byrne PM, Girgis-Gabardo A, Watson RM, Dolovich J, et al. Changes in the cellular profile of induced sputum after allergen-induced asthmatic responses. The American review of respiratory disease. 1992;145(6):1265-9.
6. Pizzichini MM, Pizzichini E, Clelland L, Efthimiadis A, Mahony J, Dolovich J, et al. Sputum in severe exacerbations of asthma: kinetics of inflammatory indices after prednisone treatment. American journal of respiratory and critical care medicine. 1997;155(5):1501-8.
7. Silkoff PE, McClean P, Spino M, Erlich L, Slutsky AS, Zamel N. Dose-response relationship and reproducibility of the fall in exhaled nitric oxide after inhaled beclomethasone dipropionate therapy in asthma patients. Chest. 2001;119(5):1322-8.
8. Wagener AH, de Nijs SB, Lutter R, Sousa AR, Weersink EJ, Bel EH, et al. External

validation of blood eosinophils, FE(NO) and serum periostin as surrogates for sputum eosinophils in asthma. Thorax. 2015;70(2):115-20.

9. Hastie AT, Moore WC, Li H, Rector BM, Ortega VE, Pascual RM, et al. Biomarker surrogates do not accurately predict sputum eosinophil and neutrophil percentages in asthmatic subjects. The Journal of allergy and clinical immunology. 2013;132(1):72-80.

10. Schleich FN, Manise M, Sele J, Henket M, Seidel L, Louis R. Distribution of sputum cellular phenotype in a large asthma cohort: predicting factors for eosinophilic vs neutrophilic inflammation. BMC pulmonary medicine. 2013;13:11.

11. Kim CK. Eosinophil-derived neurotoxin: a novel biomarker for diagnosis and monitoring of asthma. Korean journal of pediatrics. 2013;56(1):8-12.

12. Taniuchi S, Chihara J, Kojima T, Yamamoto A, Sasai M, Kobayashi Y. Serum eosinophil derived neurotoxin may reflect more strongly disease severity in childhood atopic dermatitis than eosinophil cationic protein. Journal of dermatological science. 2001;26(1):79-82.

13. Chung KF, Wenzel SE, Brozek JL, Bush A, Castro M, Sterk PJ, et al. International ERS/ATS guidelines on definition, evaluation and treatment of severe asthma. The European respiratory journal. 2014;43(2):343-73.

14. Kim SH, Yang EM, Lee HN, Choi GS, Ye YM, Park HS. Association of the CCR3 gene polymorphism with aspirin exacerbated respiratory disease. Respiratory medicine. 2010;104(5):626-32.

15. Corren J, Lemanske RF, Hanania NA, Korenblat PE, Parsey MV, Arron JR, et al. Lebrikizumab treatment in adults with asthma. The New England journal of medicine. 2011;365(12):1088-98.

16. Fajt ML, Gelhaus SL, Freeman B, Uvalle CE, Trudeau JB, Holguin F, et al. Prostaglandin D(2) pathway upregulation: relation to asthma severity, control, and TH2 inflammation. The Journal of allergy and clinical immunology. 2013;131(6):1504-12.

17. Parulekar AD, Atik MA, Hanania NA. Periostin, a novel biomarker of TH2-driven asthma. Current opinion in pulmonary medicine. 2014;20(1):60-5.

18. The ENFUMOSA cross-sectional European multicentre study of the clinical phenotype of chronic severe asthma. European Network for Understanding Mechanisms of Severe Asthma. The European respiratory journal. 2003;22(3):470-7.

19. Dolan CM, Fraher KE, Bleecker ER, Borish L, Chipps B, Hayden ML, et al. Design and baseline characteristics of the epidemiology and natural history of asthma: Outcomes and Treatment Regimens (TENOR) study: a large cohort of patients with severe or difficult-to-treat asthma. Annals of allergy, asthma & immunology : official publication of the American College of Allergy, Asthma, & Immunology. 2004;92(1):32-9.

20. Moore WC, Bleecker ER, Curran-Everett D, Erzurum SC, Ameredes BT, Bacharier L, et al. Characterization of the severe asthma phenotype by the National Heart, Lung, and Blood Institute's Severe Asthma Research Program. The Journal of allergy and clinical immunology. 2007;119(2):405-13.

21. Tran TN, Khatry DB, Ke X, Ward CK, Gossage D. High blood eosinophil count is associated with more frequent asthma attacks in asthma patients. Annals of allergy, asthma & immunology : official publication of the American College of Allergy, Asthma, & Immunology. 2014;113(1):19-24.

22. Ullmann N, Bossley CJ, Fleming L, Silvestri M, Bush A, Saglani S. Blood eosinophil counts rarely reflect airway eosinophilia in children with severe asthma. Allergy. 2013;68(3):402-6.

23. Moore WC, Meyers DA, Wenzel SE, Teague WG, Li H, Li X, et al. Identification of asthma phenotypes using cluster analysis in the Severe Asthma Research Program. American journal of respiratory and critical care medicine. 2010;181(4):315-23.

24. Wu W, Bleecker E, Moore W, Busse WW, Castro M, Chung KF, et al. Unsupervised phenotyping of Severe Asthma Research Program participants using expanded lung data. The Journal of allergy and clinical immunology. 2014;133(5):1280-8.

25. Green RH, Brightling CE, McKenna S, Hargadon B, Parker D, Bradding P, et al. Asthma exacerbations and sputum eosinophil counts: a randomised controlled trial. Lancet (London, England). 2002;360(9347):1715-21.

26. Jayaram L, Pizzichini MM, Cook RJ, Boulet LP, Lemiere C, Pizzichini E, et al. Determining asthma treatment by monitoring sputum cell counts: effect on exacerbations. The European respiratory journal. 2006;27(3):483-94.

27. Chlumsky J, Striz I, Terl M, Vondracek J. Strategy aimed at reduction of sputum eosinophils decreases exacerbation rate in patients with asthma. The Journal of international medical research. 2006;34(2):129-39.

28. Simpson JL, Scott R, Boyle MJ, Gibson PG. Inflammatory subtypes in asthma: assessment and identification using induced sputum. Respirology (Carlton, Vic). 2006;11(1):54-61.

29. Kim CK, Callaway Z, Fletcher R, Koh YY. Eosinophil-derived neurotoxin in childhood asthma: correlation with disease severity. The Journal of asthma : official journal of the Association for the Care of Asthma. 2010;47(5):568-73.

30. Gon Y, Ito R, Hattori T, Hiranuma H, Kumasawa F, Kozu Y, et al. Serum eosinophil-derived neurotoxin: correlation with persistent airflow limitation in adults with house-dust mite allergic asthma. Allergy and asthma proceedings. 2015;36(6):e113-20.

31. Ordonez CL, Shaughnessy TE, Matthay MA, Fahy JV. Increased neutrophil numbers and IL-8 levels in airway secretions in acute severe asthma: Clinical and biologic significance. American journal of respiratory and critical care medicine. 2000;161(4 Pt 1):1185-90.

32. Irvin C, Zafar I, Good J, Rollins D, Christianson C, Gorska MM, et al. Increased frequency of dual-positive TH2/TH17 cells in bronchoalveolar lavage fluid characterizes a population of patients with severe asthma. The Journal of allergy and clinical immunology. 2014;134(5):1175-86.e7.

33. Bullens DM, Truyen E, Coteur L, Dilissen E, Hellings PW, Dupont LJ, et al. IL-17 mRNA in sputum of asthmatic patients: linking T cell driven inflammation and granulocytic influx? Respiratory research. 2006;7:135.

34. Choy DF, Hart KM, Borthwick LA, Shikotra A, Nagarkar DR, Siddiqui S, et al. TH2 and TH17 inflammatory pathways are reciprocally regulated in asthma. Science translational medicine. 2015;7(301):301ra129.

35. Pham DL, Ban GY, Kim SH, Shin YS, Ye YM, Chwae YJ, et al. Neutrophil autophagy and extracellular DNA traps contribute to airway inflammation in severe asthma. Clinical and experimental allergy : journal of the British Society for Allergy and Clinical Immunology. 2017;47(1):57-70.

36. Izuhara K, Ohta S, Ono J. Using Periostin as a Biomarker in the Treatment of Asthma. Allergy, asthma & immunology research. 2016;8(6):491-8.

37. Takayama G, Arima K, Kanaji T, Toda S, Tanaka H, Shoji S, et al. Periostin: a novel component of subepithelial fibrosis of bronchial asthma downstream of IL-4 and IL-13

signals. The Journal of allergy and clinical immunology. 2006;118(1):98-104.

38. Sidhu SS, Yuan S, Innes AL, Kerr S, Woodruff PG, Hou L, et al. Roles of epithelial cell-derived periostin in TGF-beta activation, collagen production, and collagen gel elasticity in asthma. Proceedings of the National Academy of Sciences of the United States of America. 2010;107(32):14170-5.

39. Izuhara K, Nunomura S, Nanri Y, Ogawa M, Ono J, Mitamura Y, et al. Periostin in inflammation and allergy. Cellular and molecular life sciences : CMLS. 2017;74(23):4293-303.

40. Song JS, You JS, Jeong SI, Yang S, Hwang IT, Im YG, et al. Serum periostin levels correlate with airway hyper-responsiveness to methacholine and mannitol in children with asthma. Allergy. 2015;70(6):674-81.

– 국문요약 –

성인 중증천식 환자에서 생체지표로서 혈청 EDN 수치의 유용성

목적: 중증천식은 다양한 병태생리학적 기전이 혼재하는 난치성 질환으로, 중증천식의 발생에 호산구성 염증이 중요한 기여를 한다고 생각된다. 하지만, 중증천식에서 호산구성 염증을 예측할 수 있는 정립된 생체지표는 아직 발견되지 않았다. 중증천식과 비중증천식 환자에서 임상적 지표, 혈청 EDN 수치 및 다른 호산구성 생체 지표의 수치를 비교 분석하여 이러한 호산구성 생체 지표가 중증천식에서의 호산구성 염증을 예측 가능한지 알아보고자 하였다.

방법: 총 환자 중 235명의 중증천식 환자와 898명의 비중증천식 환자가 연구에 포함되었으며, 혈청 EDN 및 혈청 periostin을 측정 및 비교분석하였다. 또한, 각각의 호산구성 생체지표들과 임상적 지표들 사이의 연관성도 분석하였다.

결과: 중증천식 환자의 나이가 더 많고 천식의 유병기간도 길었으며. 천식 환자의 기저 FEV_1 및 메타콜린 PC_{20} 값이 더 낮게 측정되었다. 말초혈액 호산구 수, 객담의 호산구 비율, 혈청 EDN 수치와 혈청 periostin 수치는 중증천식 환자군이 비중증천식 환자군에 비해 유의하게 높았다. 전체 천식 환자군에서 혈청 EDN 수치는 말초혈액 호산구 수와 혈청 periostin과 양의 상관관계를 보였다.

결론: 이 연구는 혈청 EDN 수치가 중증천식에서 호산구성 염증을 예측 가능한 생체 지표로 사용될 수 있음을 제시하였다.

핵심어: eosinophil-derived neurotoxin, 중증천식, 생체지표