

ORIGINAL ARTICLE

Relation between Total Tear IgE and Severity of Acute Seasonal Allergic Conjunctivitis

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ABSTRACT

Purpose: Measurement of total tear immunoglobulin E (IgE) is useful for the diagnosis of allergic conjunctivitis, but it is still unknown whether measurement of total tear IgE is useful for assessment of the severity of allergic conjunctivitis. We evaluated the relation between the total IgE level in tears and objective signs of allergic conjunctivitis.

Methods: A prospective, nonrandomized, and cross-sectional study was conducted in 84 patients with allergic conjunctivitis (allergic group) and 80 age- and sex-matched healthy control subjects (control group). The total tear IgE score was obtained with the Allerwatch® test (0, 1, and 2), and ten severity scores (0, 1, 2, 3) were determined for objective ocular findings of the palpebral and bulbar conjunctiva, as well as limbal and corneal lesions.

Results: The scores for total tear IgE and each of the objective ocular findings were higher in the allergic group than in the control group (all $p < 0.01$). The total tear IgE score was correlated with the severity of each clinical feature of allergic conjunctivitis ($p < 0.01$), except for corneal epithelial damage in the allergic group. Multivariate analysis revealed that the severity of giant papillae was the most important indicator of the total tear IgE score (odds ratio = 1.33, $p < 0.00001$).

Conclusion: The total tear IgE score was correlated with the scores for objective signs of allergic conjunctivitis. This rapid test for total IgE in tear fluid is easy to perform and could be used to assess the severity of allergic conjunctivitis on an outpatient basis.

Keywords: Allergic conjunctivitis, Allerwatch, Immunochromatography, Objective signs, Tear, Total IgE

INTRODUCTION

The prevalence of allergic conjunctivitis is increasing along with changes of environmental factors, improvement in its diagnosis, and better understanding of the disease. Detection and quantification of total and specific immunoglobulin E (IgE) antibodies in the serum is currently the standard serological method for diagnosis of allergic conjunctivitis.^{1,2} Measurement of total IgE in tear fluid is also useful for diagnosing allergic conjunctivitis.^{3–9}

Recently, a new commercial kit (Allerwatch®, Hitachi Chemical Co., Ltd., Tokyo, Japan; and Wakamoto Pharmaceutical Co., Ltd., Tokyo, Japan) has been developed and released in Japan that provides a rapid immunoassay for total tear IgE. We reported that the

positive rate with this immunochromatography assay was 92.5% to 100.0% in patients with moderate to severe seasonal allergic conjunctivitis.^{9–11} For clinical evaluation of the severity of allergic conjunctivitis, conventional slit-lamp examination is most important, and the severity of allergic conjunctivitis has been determined from objective ocular findings on slit-lamp examination in various studies.^{12–19} Although the total IgE level in tear fluid is also a useful diagnostic indicator for allergic conjunctivitis, it is still unknown whether measurement of total tear IgE is useful for assessment of the severity of allergic conjunctivitis. Accordingly, the objectives of the present study were as follows: (1) to investigate the relation between the total tear IgE level and objective signs for allergic conjunctivitis; and (2) to assess the clinical value

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of total tear IgE for predicting the severity of allergic conjunctivitis.

MATERIALS AND METHODS

Study Design

This was a prospective, nonrandomized, and cross-sectional case study conducted at the university hospital of University of Tokyo Graduate School of Medicine and its affiliated hospitals. The study was performed in accordance with the Helsinki Declaration of 1975 and its 1983 revision. Institutional Review Board approval was obtained and informed consent was also obtained from each subject.

Subjects

All subjects had a Schirmer 1 test score greater than 15 mm and normal meibomian glands. None of them had been treated with topical or systemic drugs during the preceding 6 weeks. We excluded patients with dry eyes, patients with atopic blepharoconjunctivitis (ABC), atopic keratoconjunctivitis (AKC), or vernal keratoconjunctivitis, patients wearing contact lenses and those with a history of cataract surgery, corneal refractive surgery, or infectious conjunctivitis. Patients older than 40 years were excluded because of the higher prevalence of ABC and AKC.

Two groups were enrolled in this study (Table 1). The allergic group consisted of 84 outpatients suffering from acute seasonal allergic conjunctivitis, who were treated at our hospital and its affiliated hospitals between January and December 2009 (42 males and 42 females with a mean age of 20.3 ± 0.5 years; range: 5–39 years). In addition, we enrolled an age- and sex-matched control group, comprising 80 healthy non-smoking subjects with no history of allergic diseases and negative skin-prick test results for cedar pollen, cat dander, house dust, and *Dermatophagoides farinae* (1:20 wt/vol; Torii Pharmaceutical Co., Ltd., Tokyo, Japan) (39 males and 41 females with a mean age of 21.2 ± 0.5 years; range: 7–38 years). The control group was selected from among patients attending our outpatient clinic for eye screening tests (Table 1).

Definition of Allergic Conjunctivitis

Allergic conjunctivitis was diagnosed from the findings on slit lamp examination based on detection of features such as conjunctival hyperemia, follicles, and papillae, as well as from symptoms such as ocular itching and tearing in patients without proliferative lesions. The diagnosis was made by a single ophthalmologist (T.M.) according to published guidelines for the diagnosis and treatment of conjunctivitis.²⁰

TABLE 1 Clinical Profile of the Subjects and Results of the Allerwatch® test the Control group and the Allergic group.

Parameters	Control group	Allergic group	p value
Number of patients	80	84	
Age (years)	21.2 ± 0.5	20.3 ± 0.5	NS*
Male/Female	39/41	42/42	NS**
Allerwatch® test			
Test positive	2	72	
Test negative	78	13	<0.00001***
Sensitivity	84.5%		
Specificity	97.5%		
False negative rate	15.5%		
False positive rate	2.5%		
Positive predictive value	97.3%		
Negative predictive value	85.7%		
Positive likelihood ratio	33.8		

Comparisons between the control group and the allergic group are shown. Data are the number of patients or mean $\pm$ standard deviation. NS = not significant, *Unpaired *t* test, ** χ^2 test, ***Fisher's exact test.

The ocular findings on slit-lamp examination were scored at each patient's first visit to our clinic. Objective changes of the palpebral conjunctiva (hyperemia, edema, follicles, papillae, and giant papillae), bulbar conjunctiva (hyperemia and chemosis), and limbus (Trantas' dot and edema), as well as corneal involvement, were classified by using four grades (0 = Normal, 1+ = Mild, 2+ = Moderate, or 3+ = Severe; Table 1) as described elsewhere.^{15,18,21} In addition, the total objective score for allergic conjunctivitis (maximum of 30 points) was calculated from the above 10 findings.¹⁸ Additionally, we divided patients into five separate groups: grade 1 (total symptom score: 0–5), grade 2 (5–10), grade 3 (11–15), grade 4 (16–20), and grade 5 (21–30) according to their total symptom score. Although data were obtained for both eyes, only data from the right eye were used for analysis.

Determination of Total IgE in Tear Fluid

All patients suffered from acute seasonal allergic conjunctivitis and we obtained tear samples when the patients were symptomatic. Total IgE was determined with the Allerwatch® test according to the manufacturer's instructions. In brief, the filter strip was placed at the lower fornix of the right eye, and was removed when the tear fluid had wetted the strip to the control line. Then the strip was immersed in 100 μ L of reaction buffer in a 2.0 mL Eppendorff tube for 10 min at room temperature. The primary antibody was gold-labelled anti-human IgE. After this antibody bound to IgE in the tear fluid impregnating the test strip, it was captured by mouse anti-human IgE antibody

immobilized at the test line on the strip. Gold-labelled anti-human IgE was also immobilized as a control line on each strip. The intensity of red color developing at the test line was dependent on the total IgE content of the sample. At normal IgE levels (<2.0 KU/L), no line developed, as stated in the manufacturer's instructions. The results of the test were classified according to the following three grades: grade 0 = no detectable IgE (no line), grade 1 = a low total IgE level (test line weaker than the control line), and grade 2 = a high total IgE level (test line similar to or stronger than the control line). Classification was done by naked eye observation and then confirmed by using Scion Image software (Scion Corporation, Frederick, MD). The quantification measure was defined as the ratio between density values of a central area of 1.0 mm^2 in the test line versus the same area in the control line using the Scion Image. The three grades were defined as follows: Grade 0: less than 0.2 times control level; Grade 1: 0.2 to 1 times control level; and Grade 2: greater than one times control level.

Statistical Analysis

Mean values were compared with the unpaired t test. Frequency analysis was performed by using the χ^2 test or Fisher's exact test. Differences of semiquantitative measurements were examined by the two-tailed Mann-Whitney U test for two groups and the Kruskal-Wallis 1-way ANOVA by ranks for five groups. The sensitivity, specificity, and positive and negative predictive values were calculated with standard formulae. Relations among variables were investigated by calculating Pearson's correlation coefficients. Factors associated with the total tear IgE score were investigated by multivariate logistic regression analysis, with explanatory variables including various scores of objective ocular signs for allergic conjunctivitis. The level of significance was set at $p < 0.05$. Calculations were done using the Stat View statistical software package (Abacus Concepts, Berkeley, CA, USA).

RESULTS

The Allerwatch® test was positive in 72 of the 84 patients with allergic conjunctivitis (85.7%) and in two of the 80 controls (2.5%, Table 1). There was a significant difference of the positivity rate ($\chi^2 = 111.62$, degree of freedom [df] = 1, $p < 0.00001$, Fisher's exact test) and the grade (1.37 ± 0.74 vs. 0.03 ± 0.16 , $p < 0.00001$, Mann-Whitney U test) between the allergic group and the control group (Figure 1). The sensitivity, specificity, positive and negative predictive values, and positive likelihood ratio are shown in Table 1. The score for each objective ocular finding and the total score for allergic conjunctivitis were all higher in the allergic group than in the control group

(all $p < 0.01$, two-tailed Mann-Whitney U test, Figures 2 and 3).

The total tear IgE score was correlated with the severity of each indicator of allergic conjunctivitis ($p < 0.01$), except for corneal epithelial damage ($r = 0.172$, $p = 0.05935$, Pearson's correlation coefficient, Table 3). The strongest correlation was found between the total tear IgE score and the total score for allergic conjunctivitis ($r = 0.517$, $p < 0.00001$), followed by a correlation with giant papillae ($r = 0.437$, $p = 0.00002$, Pearson's correlation coefficient, Table 3). There was a significant difference of the total tear IgE score among the five groups divided by the total

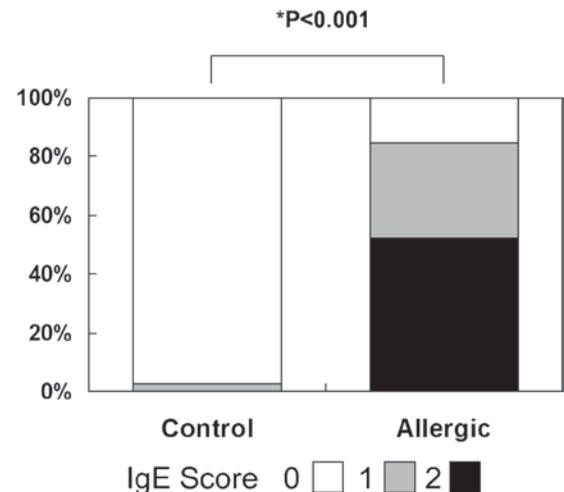


FIGURE 1 Comparison of total IgE scores between the control group and the allergic group. The total IgE score was graded as described in Methods. *Two-tailed Mann-Whitney U test.

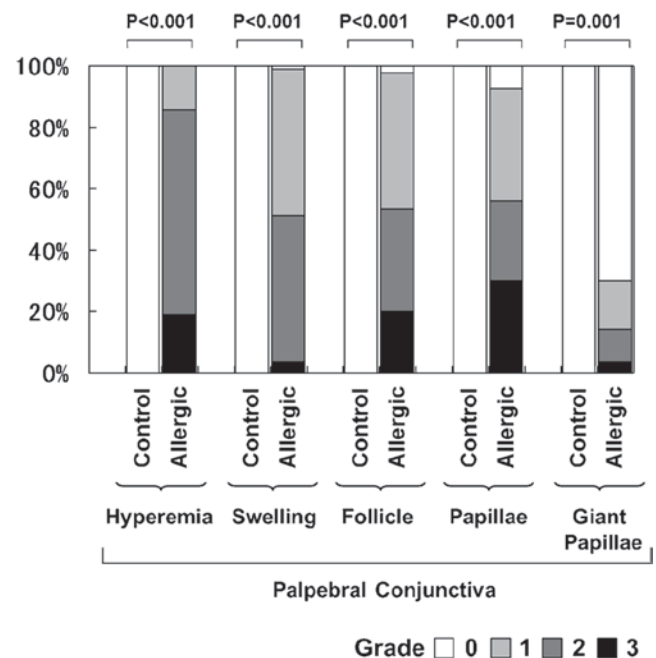


FIGURE 2 Comparison of the scores for findings of the palpebral conjunctiva between the control group ($n = 80$) and the allergic group ($n = 84$). The scores for these ocular findings were graded as described in Methods and Table 2. *Two-tailed Mann-Whitney U test.

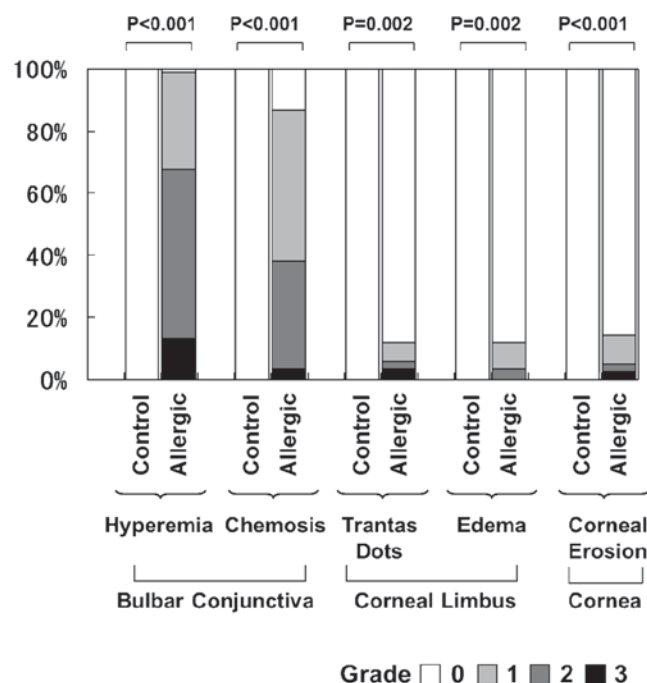


FIGURE 3 Comparison of the scores for findings of the bulbar conjunctiva, limbus, and cornea between the control group and the allergic group. The scores for the ocular findings were graded as described in "Methods" and Table 2. *Two-tailed Mann-Whitney U test.

symptom score ($p = 0.00008$, The Kruskal–Wallis test) as shown in Figure 4.

Multivariate logistic regression analysis revealed that the features of allergic conjunctivitis showed a strong association with the total tear IgE score were giant papillae (odds ratio [OR] = 1.33, confidence interval [CI] = 0.27–0.30, $p < 0.00001$) and bulbar conjunctival edema (OR = 1.30, CI = 0.25 to 0.28, $p < 0.00001$, Table 3).

DISCUSSION

This study revealed that the total tear IgE score was higher in the allergic group than in the control group. This score was also correlated with the severity of various objective features of allergic conjunctivitis. Multivariate analysis demonstrated that the presence of giant papillae and conjunctival edema were most strongly associated with the total tear IgE score. These results suggest that the total tear IgE level increases along with the severity of allergic conjunctivitis.

Although we believe the measurement of total IgE level in tear fluid is useful in the diagnosis of allergic conjunctivitis, little attention has been given to the relationships between total tear IgE level and the severity of allergic conjunctivitis. Our results showed positive correlation between the total tear IgE score and the severity of allergic conjunctivitis. Additionally, several research groups have reported a significant

TABLE 2 Grading System for Objective Clinical Signs of Allergic Conjunctivitis.*

Signs	Score
Palpebral conjunctiva	
Hyperemia	0 = None 1 = Dilatation of several vessels 2 = Dilatation of many vessels 3 = Impossible to distinguish individual blood vessels
Edema	0 = None 1 = Slight edema 2 = Diffuse edema with opacity 3 = Severe: Diffuse edema with opacity
Follicles	0 = None 1 = 1–9 follicles 2 = 10–19 follicles 3 = 20 or more follicles
Papillae	0 = None 1 = Papillae size: 0.1–0.2 mm 2 = Papillae size: 0.3–0.5 mm 3 = Papillae size: 0.6 mm or more
Giant papillae (papillae size ≥ 1 mm)	0 = None 1 = Flat papillae 2 = Elevated papillae in $<1/2$ of the upper palpebral conjunctiva 3 = Elevated papillae in $1/2$ or more of the upper palpebral conjunctiva
Bulbar conjunctiva	
Hyperemia	0 = None 1 = Dilatation of several vessels 2 = Dilatation of many vessels 3 = Diffuse dilated blood vessels over the entire bulbar conjunctiva
Edema	0 = None 1 = Localized edema 2 = Thinner diffuse edema 3 = Bullous edema
Limbus	
Trantas' dots	0 = None 1 = 1–4 dots 2 = 5–8 dots 3 = 9 or more dots
Swelling	0 = None 1 = Found in $<1/3$ of the limbal circumference 2 = Found in $1/3$ to $<2/3$ of the limbal circumference 3 = Found in $2/3$ or more of the limbal circumference
Cornea	
Corneal epithelial signs	0 = None 1 = Superficial punctate keratitis 2 = Exfoliation superficial punctate keratitis 3 = Shield ulcer or corneal erosion

*Uchio E and associates.¹⁸

TABLE 3 Correlation Coefficients for Comparisons of Total IgE and Various Allergic Scores and Multivariate Odds Ratios and 95% Confidence Intervals for Total Tear IgE in the Allergic Group ($n = 84$).

Variables	Correlation coefficients		Multivariate analysis		
	R	<i>p</i> value	OR	(95% CI)	<i>p</i> value
<i>Palpebral conjunctiva</i>					
Hyperemia	0.436	0.00002	1.06	(0.04–0.08)	<0.00001
Edema	0.507	<0.00001	0.93	(–0.09 to –0.06)	<0.00001
Follicles	0.376	0.00021	0.96	(–0.04 to –0.02)	<0.00001
Papillae	0.418	0.00004	1.07	(0.05–0.08)	<0.00001
Giant papillae	0.437	0.00002	1.33	(0.27–0.30)	<0.00001
<i>Bulbar conjunctiva</i>					
Hyperemia	0.321	0.00146	1.04	(0.02–0.05)	<0.00001
Edema	0.356	0.00045	1.30	(0.25–0.28)	<0.00001
<i>Limbus</i>					
Trantas' dots	0.255	0.00956	1.06	(0.03–0.09)	0.00011
Swelling	0.259	0.00871	0.70	(–0.40 – –0.33)	<0.00001
<i>Cornea</i>					
Epithelial damage	0.172	0.05935	0.87	(–0.16 – –0.13)	<0.00001
<i>Total Score</i>	0.517	<0.00001	–	–	–

R = Two-tailed Pearson's correlation coefficients were calculated to assess the association between the total tear IgE score and various features of allergic conjunctivitis; OR = Odds ratio; CI = Confidence interval.

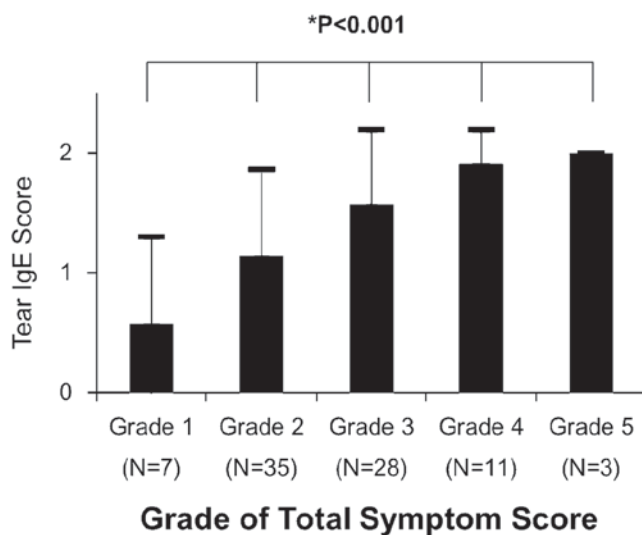


FIGURE 4 Comparison of the mean total IgE scores among five groups of different degrees of total symptom score. Patients were divided into five separate groups according to total symptom score as described in Methods. Data are the mean $\pm$ SD. *The Kruskal–Wallis test.

correlation between symptoms of ocular allergy and both total IgE in tears and allergen-specific IgE in tears, whereas only a poor correlation has been found between specific and/or total IgE in serum and the manifestations of ocular allergy.^{5, 22–24} We also examined the relationship between total tear IgE and serum IgE in patients with allergic conjunctivitis, and there was a moderate correlation between the total tear IgE score and log (total serum IgE).¹⁰ These reports and our results suggest local production of IgE is the chief contributor to the manifestations of allergic conjunctivitis. Therefore, measurement of locally produced total tear IgE is an

efficient practical method for the clinical evaluation of allergic conjunctivitis.

In the present study, the Allerwatch® test showed a very high sensitivity (84.5%) and specificity (97.5%). With regard to another diagnosis kit for allergic conjunctivitis (Lacrytest®, ADIATEC S.A, Nantes, France), Sirbikate *et al.* reported that the sensitivity and specificity were 93.8% and 89.7%, respectively,⁶ so the present rates were similar to or higher than those reported previously.^{6,7}

Over the past 20 years, various clinical scoring systems for allergic conjunctivitis have been proposed.^{12–19} We used the latest scoring system developed in Japan.^{15,18} This system is based on objective parameters, and involves scoring ten ocular findings on a four-point scale. It is easy to use and allows reliable and reproducible evaluation of the severity of allergic conjunctivitis. On the other hand, measurement of total tear IgE is also useful for the diagnosis of allergic conjunctivitis.^{3–8} In this study, the total tear IgE score was significantly correlated with the clinical score for allergic conjunctivitis, suggesting that measurement of total tear IgE can be useful for determining the severity of allergic conjunctivitis. Giant papillae had the strongest correlation with the total IgE score. Donshik and associates reported that increased tear fluid IgE levels were found in the more symptomatic eyes of patients with giant papillary conjunctivitis.²⁵ Studies using transferrin as a marker for the leakage of serum proteins into the tears have shown that local production is responsible for increased tear fluid levels of IgE.²⁵ Increased vascular permeability of the palpebral conjunctiva may contribute directly to the local production of IgE that is released into the tears.

Immunochromatographic analysis of total IgE in tear fluid has several advantages over enzyme-linked

immunosorbent assay as a standard method of analysis. First, less time is required for measurement of IgE levels than when using standard assays. We could measure the total IgE level in 10 min with the present immunochromatography assay. This could be of considerable benefit to patients, because allergic conjunctivitis can be diagnosed during the first visit. Second, this assay has potential as a noninvasive test for diagnosis of allergic conjunctivitis by simply touching a piece of filter paper to the lower fornix of the conjunctiva. Therefore, immunochromatography provides a rapid, sterile, and noninvasive method for confirmation of the diagnosis of allergic conjunctivitis.

The major disadvantage is that this method cannot be used in patients with dry eyes. In fact, it is difficult to measure total IgE levels accurately in such patients. Fujishima and associates reported that patients with allergic conjunctivitis who were negative for serum specific IgE had lower Schirmer's test scores, a lower tear clearance, a lower tear function index, and lower serum total IgE than serum IgE-positive patients with allergic conjunctivitis.²⁶ Patients with dry eyes and allergic conjunctivitis sometimes have similar symptoms. Therefore, dry eyes may affect both the measurement of total tear IgE and the scores for objective signs of allergic conjunctivitis. Second, the IgE level detected by this kit of immunochromatographic assay was a considerably rough estimation. Enzyme-linked immunosorbent assay (ELISA) may be of value for a high-quality study to determine tear IgE concentration, and then to analyze the relationship with ocular signs and the severity of allergic conjunctivitis. Although the reproducibility of the assay is reliable as described in the manufacturer's protocol, further evaluation of the accuracy of the assay using other quantitative methods such as ELISA and Western blot test is needed. Third, although detection of eosinophils in body fluids/tissues is used for the diagnosis of allergic conditions,²⁷⁻²⁹ we did not investigate eosinophils in the collected samples. Detection of eosinophils is a complicated procedure and the detection rate is low, being about 60% in conjunctival scrapings from patients with mild allergic conditions.²⁷ Therefore, rapid measurement of local IgE is preferable for diagnosis of allergic diseases instead of the traditional eosinophil count. Fourth, we excluded patients with atopic keratoconjunctivitis from the study because this research focused on seasonal allergic conjunctivitis. Inada and associates reported that the total IgE score was significantly higher in atopic and vernal keratoconjunctivitis than in seasonal and perennial allergic conjunctivitis.⁸ In the future, we will investigate the relationship between the total IgE level in tears and the specific serum IgE in patients with AKC.

In conclusion, this study demonstrated that the total tear IgE score was closely correlated with the severity of objective signs of allergic conjunctivitis in patients with allergic conjunctivitis. A new rapid

immunochromatography test for measurement of total IgE in tear fluid may be useful for determining the severity of allergic conjunctivitis as a rapid point-of-care assay.

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REFERENCES

- [1] Mimura T, Amano S, Funatsu H *et al.* Correlations between allergen-specific IgE serum levels in patients with allergic conjunctivitis in spring. *Ocul Immunol Inflamm* 2004;12:45-51.
- [2] Mimura T, Yamagami S, Amano S *et al.* Allergens in Japanese patients with allergic conjunctivitis in autumn. *Eye (Lond)* 2005;19:995-999.
- [3] Ballou M, Mendelson L. Specific immunoglobulin E antibodies in tear secretions of patients with vernal conjunctivitis. *J Allergy Clin Immunol* 1980;66:112-118.
- [4] Liotet S, Warnet VN, Arrata M. Lacrimal immunoglobulin E and allergic conjunctivitis. *Ophthalmologica* 1983;186:31-34.
- [5] Nomura K, Takamura E. Tear IgE concentrations in allergic conjunctivitis. *Eye (Lond)* 1998;12 (Pt 2):296-298.
- [6] Sirbikaite L, Ehlers N. The use of Lacrytest in ocular allergy. *Acta Ophthalmol Scand* 2007;85:117.
- [7] Monzón S, Arrondo E, Bartra J, *et al.* Conjunctivitis and total IgE in lacrimal fluid: Lacrytest screening. *Journal of Allergy* 2009;54.
- [8] Inada N, Shoji J, Kato H, Kiely S, Mulyanto, Sawa M. Clinical evaluation of total IgE in tears of patients with allergic conjunctivitis disease using a novel application of the immunochromatography method. *Allergol Int* 2009;58:585-589.
- [9] Mimura FT, Usui T, Mori M *et al.* Rapid immunochromatography of total tear immunoglobulin E in allergic conjunctivitis with Allerwatch. *J Investig Allergol Clin Immunol* 2010;20:627-628.
- [10] Mimura T, Usui T, Mori M *et al.* Relationship between total tear and serum IgE in allergic conjunctivitis. *Int Arch Allergy Immunol* 2011;154:349-352.
- [11] Mimura T, Usui T, Mori M *et al.* Relation between total tear IgE and specific serum IgE in seasonal allergic conjunctivitis. *Cornea* 2011;30:790-795.
- [12] Abelson MB, Chambers WA, Smith LM. Conjunctival allergen challenge. A clinical approach to studying allergic conjunctivitis. *Arch Ophthalmol* 1990;108:84-88.
- [13] Leonardi A, Borghesan F, Avarello A, Plebani M, Secchi AG. Effect of lodoxamide and disodium cromoglycate on tear eosinophil cationic protein in vernal keratoconjunctivitis. *Br J Ophthalmol* 1997;81:23-26.
- [14] Brémond-Cignac D. The clinical spectrum of ocular allergy. *Curr Allergy Asthma Rep* 2002;2:321-324.
- [15] Japanese Ocular Allergology Study Group. Guidelines for the clinical management of allergic conjunctival diseases. The Japanese Ophthalmological Society. *Nippon Ganka Gakkai Zasshi* 2006;110:103-140.
- [16] Calonge M, Herreras JM. Clinical grading of atopic keratoconjunctivitis. *Curr Opin Allergy Clin Immunol* 2007;7:442-445.
- [17] Bonini S, Sacchetti M, Mantelli F, Lambiase A. Clinical grading of vernal keratoconjunctivitis. *Curr Opin Allergy Clin Immunol* 2007;7:436-441.
- [18] Uchio E, Kimura R, Migita H, Kozawa M, Kadonosono K. Demographic aspects of allergic ocular diseases and

- evaluation of new criteria for clinical assessment of ocular allergy. *Graefes Arch Clin Exp Ophthalmol* 2008;246:291–296.
- [19] Shoji J, Inada N, Sawa M. Evaluation of novel scoring system named 5-5-5 exacerbation grading scale for allergic conjunctivitis disease. *Allergol Int* 2009;58:591–597.
- [20] Ben Ezra D. Guidelines on the diagnosis and treatment of conjunctivitis. *Ocul Immunol Inflam* 1994;2 (Suppl):17–26.
- [21] Ohashi Y, Ebihara N, Fujishima H *et al.* A randomized, placebo-controlled clinical trial of tacrolimus ophthalmic suspension 0.1% in severe allergic conjunctivitis. *J Ocul Pharmacol Ther* 2010;26:165–174.
- [22] Hoffmann-Sommergruber K, Ferreira ED, Ebner C *et al.* Detection of allergen-specific IgE in tears of grass pollen-allergic patients with allergic rhinoconjunctivitis. *Clin Exp Allergy* 1996;26:79–87.
- [23] Batellier L, Poilane C, Rault J, Chaumeil C, Scat Y. [Measurement of total IgE in tears: the adaptation of an immunoenzyme technique and the value of investigating locally produced IgE in the diagnosis of chronic conjunctivitis]. *Ann Biol Clin (Paris)* 1999;57:469–473.
- [24] Shoji J, Kato H, Kitazawa M, Inada N, Sawa M. Evaluation of staphylococcal enterotoxin-specific IgE antibody in tears in allergic keratoconjunctival disorders. *Jpn J Ophthalmol* 2003;47:609–611.
- [25] Donshik PC, Ballow M. Tear immunoglobulins in giant papillary conjunctivitis induced by contact lenses. *Am J Ophthalmol* 1983;96:460–466.
- [26] Fujishima H, Toda I, Shimazaki J, Tsubota K. Allergic conjunctivitis and dry eye. *Br J Ophthalmol* 1996;80:994–997.
- [27] Abelson MB, Madiwale N, Weston JH. Conjunctival eosinophils in allergic ocular disease. *Arch Ophthalmol* 1983;101:555–556.
- [28] Leonardi A, Borghesan F, Faggian D, Depaoli M, Secchi AG, Plebani M. Tear and serum soluble leukocyte activation markers in conjunctival allergic diseases. *Am J Ophthalmol* 2000;129:151–158.
- [29] Wakamatsu TH, Tanaka M, Satake Y *et al.* Eosinophil cationic protein as a marker for assessing the efficacy of tacrolimus ophthalmic solution in the treatment of atopic keratoconjunctivitis. *Mol Vis* 2011;17:932–938.