Optimal Allergens for Diagnostic Testing and Biomarkers of the Response to Allergen Immunotherapy

Jeju National University School of Medicine, Jeju, Korea

Jaechun Lee

Introduction

Aeroallergens are any airborne substance capable of binding immunoglobulin E (IgE) that can trigger an allergic reaction.[1] Aeroallergens are respirable/inhalable particles to which some people become sensitized upon exposure that cause respiratory allergies, which can be manifested as allergic rhinitis, allergic conjunctivitis, or atopic asthma.

The detection of causative aeroallergens is essential for accurate diagnosis and proper management. Avoidance of aeroallergens is one strategy, but it is important to identify the specific aeroallergens to avoid at the beginning of treatment. Identification of aeroallergens provides some clues about the expected disease severity of a patient receiving pharmacological therapy. As the only disease-modifying therapy at present, allergen-specific immunotherapy (SIT) requires the identification of allergens for targeting before the start of therapy.

SIT is approved for the treatment of respiratory allergic disorders and is based mainly on empirically proven traditional treatments. A deeper understanding of the mechanisms responsible for allergic disorders may help to identify the direct link between an individual immune measurement (biomarker) and the clinical manifestation. The recent EAACI Position Paper lists biomarkers for monitoring the clinical efficacy of SIT,[2] and forms the basis of this brief review.

1. Optimal allergens for diagnostic testing

In clinical practice, allergic sensitization is detected by *in vivo* (provocation test and skin test) and *in vitro* (serum specific IgE) methods. Some environmental particles from plant pollen, fungi, mites, mammals, insects, and food

are known as aeroallergens. Hundreds of allergens in each category have been identified as causes of respiratory allergies and have been catalogued in a database (www.allergome.org). Of the myriad aeroallergens identified, the essential question is which allergens should be tested in clinical practice, and the answer to this question requires consideration of many factors.

Aeroallergen density

The density of aeroallergens to which patients are exposed in their living and working environment should be considered. The individual pollen count is the main starting consideration. Sensitization to aeroallergens depends mainly on the aeroallergen density powered by exposure duration. As an example, Japanese cedar (JC) pollinosis is a national affliction in Japan during the flowering season of early spiring.[3] By contrast, JC pollinosis is not common among Koreans because JC pollen is not found in Korea except for Jeju Island, as shown in the pollen calendar.[4]

Sensitization rate

Not all particles in air are aeroallergens. Aeroallergens are defined by their sensitization characteristics in terms of the specific IgE response elicited in humans. Sensitization may differ according to ethnicity and can vary geographically within a specific ethnic group.[5,6] Each person's microenvironment may also have implications for sensitization.[7] The sensitization rates in a community or in a diseased group should be considered so that commonly sensitized aeroallergens are included in diagnostic testing.

Disease-related aeroallergens

Sensitization to aeroallergens is common, but sensitization does not necessarily translate to a clinically relevant respiratory allergy. In sensitized people, the clinical relevance can be shown by either strong circumstantial evidence of the response to allergen exposure or direct aeroallergen challenge.8 Clinically relevant sensitization rates for some aeroallergens can differ markedly between regions.[9]

Cost-effectiveness

A diagnostic test that includes more aeroallergens is more likely to provide an accurate diagnosis of respiratory allergies. Polysensitization is defined as the allergic sensitization to two or more allergens, which is highly prevalent in people with respiratory allergies.[10] However, not all allergens associated with polysensitization are clinically relevant allergens.

The minimum screening panel for the diagnosis of allergic rhinitis has been investigated in China. Eight allergens (Der f, Der p, mugwort, *Blatella* spp., hazel, goosefoot, *Penicillium notatum*, and animal dander) are sufficient for identifying patients with allergic rhinitis in central China.[11] A panel of 10 commonly sensitized aeroallergens has been suggested in Korea. Except for Chungnam and Jeju, the 10 aeroallergens account for the causes of allergy in

more than 90% of sensitized Koreans.[5]

Availability

Many standardized aeroallergens are commercially available from several companies, although the content of aeroallergens may differ between manufacturers. The allergenicity of standardized aeroallergens may also differ according to where the allergens originated and whether by-products such as endotoxins have been efficiently removed.[12]

2. Biomarkers of the response to allergen immunotherapy

Biomarkers can be used in the quantitative measurement of the response to allergens and for the diagnosis and assessment of the stage of allergy, prediction of clinical outcomes, and monitoring of treatment.[13] SIT is effective in reducing the severity of symptoms and skin responses, need for rescue medicine, and provocative allergen threshold.*14* In addition to these therapeutic effects, the disease-modulating effects and long-term benefits after cessation of SIT are beneficial in the management of respiratory allergies. Biomarkers are needed to identify those likely to benefit from SIT and when to start and stop SIT, to predict relapses, and to determine when to boost SIT.[2] In spite of no validated candidate predictive of the treatment response, biomarkers to assess the objective treatment effect are evidenced.

The EAACI Position Paper on biomarkers for monitoring the clinical efficacy of SIT classifies biomarkers into seven categories: IgE, IgG subclasses, serum inhibitory activity for IgE, basophil activation, cytokines/chemokines, cellular biomarkers, and *in vivo* biomarkers. Some of these are used in clinical practice and are promising in the near future. Serial changes in these biomarkers are usually measured before, during, and after SIT.

IgE

Elevated specific IgE (sIgE) concentration in the serum and clinically relevant symptoms are the only criteria and the gold standard for deciding whether to start SIT. An increased sIgE concentration during SIT reflects adequate immunogenicity and/or allergen exposure. An elevated sIgE to total IgE ratio is a potential method for distinguishing between responders and nonresponders.

IgG subtypes

Low sIgG4 is a potential negative predictive marker. Failure of sIgG4 induction indicates poor compliance.

Serum inhibitory activity for IgE

IgE blocking factors (IgE-BF) are mainly in the form of IgA and IgG in the serum and show inhibitory effects by blocking allergen binding to IgE. Because of their limited availability, IgE-BF are not candidate biomarkers. The IgE-facilitated allergen binding (IgE-FAB) assay using flow cytometry is an *in vitro* model of allergen presentation

through CD23 on B cells. IgE–FAB activity differs between SIT responders and nonresponders.

Basophil activation

Basophil activation in response to sensitized allergen reflects the FcγRI–mediated response in the body. However, handling viable basophils is technically challenging, and standardization of the assay is necessary before it can be used in clinical applications.

Cytokines and chemokines

Cytokines and chemokines are locally produced, and their concentrations become diluted once in the circulatory system. These are useful only for identifying organ–specific local responses at the site of inflammation.

Cellular markers

Regulatory T cells (Treg) are involved in the Th2 to Th1 response and the change in allergen–specific B cells toward the direction of regulatory B cells. However, no specific marker of Treg is available. Instead, measuring dendritic cells (DCs) in the blood or tissues may be more promising.

In vivo biomarkers

The provocation test with allergens applied to the affected organ can be used to quantify the changes in the response from before to during and after SIT. It has been used as a biomarker of SIT with plenty of dose–response and time–course data. Skin testing, nasal or conjunctival challenge, and environmental exposure chambers (EEC) are used. EEC are useful for controlling environmental factors such as temperature, humidity, and the symptomatic variability of respiratory allergies. With standardization and clinical validation, comparison study is necessary between provocation and natural symptoms.

Conclusions

To select optimal allergens for diagnostic testing is the mainstay for accurate diagnosis and proper management. High–density aeroallergens as frequent sensitizers with clinical relevance should be considered in the practice. Biomarkers in SIT provide quantitative measurement of the response, which facilitates clinical decisions.

References

1. Alvarez-Cuesta E, Bousquet J, Canonica G, Durham S, Malling HJ, Valovirta E. Standards for practical allergen-specific immunotherapy. Allergy 2006;61:1-3.
2. Shamji M, Kappen J, Akdis M, et al. Biomarkers for monitoring clinical efficacy of allergen immunotherapy for allergic rhinoconjunctivitis and allergic asthma: an EAACI Position Paper. Allergy 2017;72:1156-73.
3. Yamada T, Saito H, Fujieda S. Present state of Japanese cedar pollinosis: the national affliction. J Allergy Clin Immunol 2014;133:632-9. e5.
4. Oh JW, Lee HB, Kang IJ, et al. The Revised Edition of Korean Calendar for Allergenic Pollens. Allergy Asthma Immunol Res 2012;4:5-11.
5. Kang MG, Kim MY, Song WJ, et al. Patterns of Inhalant Allergen Sensitization and Geographical Variation in Korean Adults: A Multicenter Retrospective Study. Allergy Asthma Immunol Res 2017;9:499-508.
6. Lee J, Lee KH, Lee HS, Hong SC, Kim JH. Japanese Cedar (*Cryptomeria japonica*) Pollinosis in Jeju, Korea: Is It Increasing? Allergy Asthma Immunol Res 2015;7:295-300.
7. Park Y-B, Mo E-K, Lee J-Y, et al. Association between pet ownership and the sensitization to pet allergens in adults with various allergic diseases. Allergy Asthma Immunol Res 2013;5:295-300.
8. Brożek JL, Bousquet J, Agache I, et al. Allergic Rhinitis and its Impact on Asthma (ARIA) guidelines—2016 revision. J Allergy Clin Immunol 2017;140:950-8.
9. Burbach GJ, Heinzerling LM, Edenharter G, et al. GA2LEN skin test study II: clinical relevance of inhalant allergen sensitizations in Europe. Allergy 2009;64:1507-15.
10. Migueres M, Dávila I, Frati F, et al. Types of sensitization to aeroallergens: definitions, prevalences and impact on the diagnosis and treatment of allergic respiratory disease. Clinical and translational allergy 2014;4:16.
11. Lou H, Ma S, Zhao Y, et al. Sensitization patterns and minimum screening panels for aeroallergens in self-reported allergic rhinitis in China. Sci Rep 2017;7:9286.
12. Jeong KY, Choi S-Y, Lee J-H, et al. Standardization of house dust mite extracts in Korea. Allergy Asthma Immunol Res 2012;4:346-50.
13. Paczesny S, Hakim FT, Pidala J, et al. National Institutes of Health Consensus Development Project on criteria for clinical trials in chronic graft-versus-host disease: III. The 2014 Biomarker Working Group report. Biol Blood Marrow Transplant 2015;21:780-92.
14. Bousquet J, Lockey R, Malling H-J. Allergen immunotherapy: therapeutic vaccines for allergic diseases A WHO position paper. J Allergy Clin Immunol 1998;102:558-62.