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RESEARCH

SPECIFIC IGE IMMUNOBLOT METHOD IN ALLERGIC RHINITIS

(IgE Spesifik Menurut Metode Imunoblot di Rinitis Alergi)

Izzuki Muhashonah[1], Aryati[1], Dwi Reno Pawarti[2], M. Robi'ul Fuadi[1], Janti Trihabsari[1]

ABSTRAK

Rinitis alergi merupakan penyakit bukan akibat non-infeksi yang ditemukan antara 10−30% penduduk dewasa dunia dan dapat menyebabkan penurunan mutu kehidupan seseorang. Rinitis alergi merupakan manifestasi alergi tipe 1 atau tipe cepat yang dimediasi oleh IgE. Pemeriksaan utama rinitis alergi adalah Skin Prick Test (SPT) dan IgE spesifik. Pemeriksaan IgE spesifik mempunyai kepekaan dan kekhasan yang menyerupai SPT, tidak memerlukan tenaga terlatih dan menyebabkan anafilaktik. Penelitian ini untuk mengetahui adakah kesesuaian nilai diagnostik IgE spesifik menurut metode imunoblot dengan SPT di pasien rinitis alergi dengan mengujinya. Rancangan penelitian adalah potong lintang yang dilakukan terhadap pasien yang datang di Unit Rawat Jalan THT-KL RSUD Dr. Soetomo pada bulan Mei sampai dengan Oktober 2014. Pasien dikelompokkan berdasarkan diagnosis rinitis alergi dan yang non-alergi dan non-infeksi serta ditetapkan secara klinis, ada riwayat alergi, pemeriksaan fisik, serta tingkat jumlah keseluruhan IgE serum dan atau eosinofil darah. Pemeriksaan SPT dilakukan dengan memakai ekstrak alergen dari Stallergens dan IgE spesifik menurut metode imunoblot memakai Foresight®. Dalam kajian ini didapatkan empat puluh tiga pasien didiagnosis rinitis akibat alergi. Hasil IgE spesifik menurut metode imunoblot positif terdapat di 36 (84%) pasien dengan pola alergen terbanyak D1/D2 29 (67%). Kepekaan dan kekhasan diagnostik IgE spesifik menurut metode imunoblot berturut-turut adalah 72,34% dan 46,15%. Kesesuaian nilai diagnostik IgE spesifik menurut metode imunoblot dengan SPT mempunyai koefisien kappa 0,158. Didasari telitian ini tidak didapatkan kesesuaian antara IgE spesifik menurut metode imunoblot dengan SPT. Di ketahui pula bahwa IgE spesifik menurut metode imunoblot dapat digunakan bersama-sama dengan SPT dalam mendiagnosis rinitis akibat alergi.

***Kata kunci**: Rinitis akibat alergi, Skin Prick Test, IgE spesifik menurut metode imunoblot*

ABSTRACT

Allergic rhinitis is a non-infectious disease found in between 10−30% of the adult population in the world and can cause a decreased quality of life. Allergic rhinitis is an allergic type manifestation of immediate type 1 mediated by IgE. The most common test for allergic rhinitis is Skin Prick Test (SPT) and specific IgE. The examination of specific IgE has a sensitivity and specificity that resembles SPT. This method does not require skilled personnel as well as cause anaphylaxis. This study aimed to know the diagnostic compatibility between specific IgE immunoblot with SPT in allergic rhinitis patients by assessing it. The study was conducted on patients at the Outpatient Clinic Department of ENT-Head and Neck from May until October 2014 by cross-sectional design. Patients were grouped as allergic rhinitis and non-allergic non-infectious rhinitis based on the clinical signs and symptoms, atopy history, physical examination, the concentration of total serum IgE and/or blood eosinophils. The skin Prick Test examination was conducted by using allergen extracts from Stallergens® and specific IgE immunoblot using Foresight®, Acon Laboratories. Based on this study, found forty-three samples which diagnosed as allergic rhinitis. Positive specific IgE immunoblot was shown in 36 (84%) patients with the highest allergen patterns D1/D2 in 29 (67%). Diagnostic sensitivity and specificity of specific IgE immunoblot were 72.34% and 46.15%, respectively. The diagnostic compatibility value of specific IgE immunoblot with SPT showed a kappa coefficient of 0.158. There was no compatibility between specific IgE immunoblot with SPT thus, specific IgE Immunoblot method and the SPT examination can be used together for allergen confirmatory and diagnosis of allergic rhinitis.

Key words: Allergic rhinitis, skin prick test, specific IgE immunoblot method

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INTRODUCTION

Rhinitis is a world health problem found in between 20–40% population of developing countries and it is increasing over years.[1] Rhinitis or inflammation disease occurred in the nose mucosa is grouped into two types: allergic and non-allergic rhinitis. The one frequently is allergic rhinitis infecting 10–20% of population. Patients suffering from allergic rhinitis increase in this modern era because of lower life quality and sleep deprivation. There are, those suspected with allergic rhinitis diagnosis visiting Outpatient Clinic Department of ENT-Head and Neck Dr. Soetomo Hospital, between 10–20 patients every month.[3]

The diagnosis of allergic rhinitis is determined by history, physical and laboratorium test covering: skin test, eosinophil, total IgE, specific IgE related to serum and nasal provocation test. Diagnosing allergic rhinitis based on Allergic Rhinitis and its Impact on Asthma World Health Organization (ARIA-WHO 2008) depends on the result of history and physical test supported by Skin Prick Tes (SPT) or specific IgE serum.[1,4] Recently, checkup for allergic rhinitis diagnosis at the most used SPT. However, there are some weaknesses using such method; for instances uncomfortable feeling of the patients, expertise necessary and high-risk of anaphylaxis.[5,6] It could not be executed to patients with severe urticaria and eczema and to those who take medical treatment for a lifetime.[7,8] Another laboratory test besides SPT is specific IgE test. The latter test is developing because it minimizes risk, is easily performed with a sensitivity and specificity similar to SPT that can at be determined as allergic diagnosis.[5,9]

The IgE test is also developing fast using several methodologies; ELISA, radioimmuno assay, chemiluminescence assay and immunoblot assay. Immunoblot test is recently popular because its sensitivity and specificity is similar to SPT and several types of allergen could be recognized. The specific IgE test based on immunoblot method could be conducted in children and patients with large eczema who are unable to take SPT test and it does not cause anaphylaxis risk.[5,9] Immunoblot method has various methods and each differ in its diagnostic value. There has not been any tests and studies about specific IgE based on immunoblot method conducted in Outpatient Clinic Department of ENT-Head and Neck Dr. Soetomo Hospital Surabaya. Therefore, this research is aimed to study the diagnostic values of specific IgE test based on immunoblot for patients with allergic rhinitis in hospital.

METHODS

This research used a cross-sectional design based on the standard of ARIA-WHO 2008 during six months, since May to October 2014. It was conducted in Outpatient Clinic Department of ENT-Head and Neck Dr. Soetomo Hospital Surabaya on those patients who had allergic rhinitis. The researchers collected the samples from all patiens diagnosed between 10-65-year old as suffering from allergic rhinitis and willingly to participate in this study. From those samples, 43 out of 60 met the criteria; while the other 17 will be as the control group.

The data of characteristic of patients consisted of age, sex, signs and symptoms, blood eosinophil and the total serum Immunoglobulin E. Based on the immunoblot method, the variable, studied were SPT results and serum allergen-specific Immunoglobulin E by immunoblot. For the SPT, the researcher used 16 kinds of allergen reagents from *Stallergenes*® with a special *lancet* from *Stallerpoint*®. The specific immunoglobulin E used 6 mL patients' serum from venous blood centrifuged at 3000 rpm for 15 minutes. In addition, its serum was kept at a stable temperature of -20^{o}C for six months which then was tested with *Foresight*®'s immunoblot reagents from *Acon Lab*. It took 2.5 hours for the testing. After that process, the test result as its research data was evaluated descriptively and analyzed based on the diagnostics of specific Immunoglobulin E.

Measuring the conformity level between specific Immunoglobulin and SPT, this research used Kappa coefficient. Kappa measurement defined the valid result will be: <0.4: bad; 0.4-0.6: fine; 0.6-0.75: satisfying and >0.75: special. Based on the immunoblot method, the specific Immunoglobulin E result was determined through the differences between allergic rhinitis and non-allergic rhinitis with Mann-Whitney U test. All the statistical analysis were processed by SPSS software.

RESULTS AND DISCUSSION

Allergic rhinitis is closely related to the quality of life of patiens; doctors need more specific consent to figure out the cause of allergy to determine suitable treatment and prevent recrudescent. Diagnosis of allergic rhinitis is based on medical check up covering: clinic, history of allergic records, physical test, eosinophil level in blood and IgE serum level and SPT. Supplementary test of SPT could help patient to detect his or her allergic rhinitis diagnosis. History and clinical symptoms test were directed to allergic

diagnosis; meanwhile SPT and laboratory tes are used to declare the existence of specific IgE antibody towards allergen within patients' skin and blood. Both tests help to identify common allergen spotted on patients' body. Allergic test result must be related to indication of: clinic, age, proper allergen exposure and the characteristics of specific test (sensitivity, exclusivity and its development).[10]

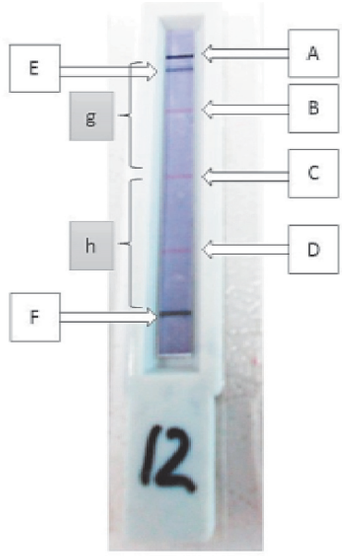

Figure 1. The characteristics of specific IgE test based on *Foresight*® immunoblot methodology **A**) Line control that has to appear in the end of each test; **B, C, D**) divider line intra-group of allergen; **E**) positive test line of allergen of specific IgE; **F**) positive test line of the increase of whole IgE level; **G**) space between line A-B and B-C: where Inhalant allergen group test lines are located; **H**) space between C-D and D-F: where Ingestan allergen group test lines are located.

The guarantee of test result determining specific IgE concentration based on Foresight® immunoblot method is written through line control in the end of every session. The comparative value is accepted if it follows the rules of exclusivity in every batch. Line control is a signifier showing that the result from immunoblot is valid and trusted; therefore, it could be used to detect reagent's damage. The result is invalid if there is no result in this research. It shows that immunoblot apparatus is well functioned. The example of the test result is shown in Figure 1.

The result of this study was reviewed by three persons in reading immunoblot line to avoid subjectivity simultaneously. Immunoblot line is interpreted 15 to maximum 30 minutes after the test. Conformity amongst the readers is summed; with results on kappa coefficient 0.722. It means that there is a good or satisfied conformity among them because they do not read different result.

The collected data shows there were no significant differences between the two groups in terms of age and sex factors; thus both were homogeneous and valid to be processed through statistical calculation. The data which contained complaints from patients included: sneezing, itchiness on the nose area and recurring cold differs on statistically calculation, however those who suffer both types of rhinitis indicate stuffed nose symptoms.

The result of testing the whole IgE serum had significant impact to determine diagnosis allergic rhinitis with r=0.405 and p=0.001. This study showed that 58% of allergic rhinitis patients experienced the increase of IgE serum (≥100 IU/mL) and 42% of them did not. Other studies conducted on children in Iran

Table 1. Characteristics of subjects based on group diagnosis

Characteristics	Group		Value *P*
	Allergic Rhinitis (n=43)	Non-Allergic Rhinitis, non-infection (n=17)	
Sex			0.843
Male	14 (32.56%)	6 (35.29%)	
Female	29 (67.44%)	11 (64.71%)	
Age (years)			
Average±SB	32.07±11.34	36.59±14.83	0.393
Grievances			
Sneezing	37 (86.05%)	7 (41.18%)	0.0001
Stuffed nose	37 (86.05%)	16 (94.12%)	0.389
Itchiness on nose area	34 (79.07%)	5 (29.41%)	0.0001
Recurring cold	38 (88.37%)	6 (35.29%)	0.0001
The whole amount of IgE serum (IU/μL)			
Average	26.,96±322.27	143.30±404.09	0.001
Median	134.40	19.90	
Eosinophil in blood			
Absolute (x10^3/μL)	0.296±0.206	0.184±151.41	0.049
Percentage (%)	3.87±2.59	2.5±2.28	0.024

proved that those with allergic rhinitis have an increase of 35% to 74% of their IgE serum level.[15]

The clinical appearance of patients with allergic rhinitis was followed by the increase of eosinophils in blood which was found to be 86.04%. It was not different with previous study held by Aryati[16] stating that excalation of eosinophil level was up to 75.64%.[16] It had a significant result on statistical calculation among allergic and non-infectious rhinitis with r=0.255 and p=0.049. This explained that eosinophil was an important factor in allergic reaction.[12] Eosinophils triggered allergy which was found in respiratory tract of patients. A previous study stated that declined eosinophils was related to disease improvement.[17]

SPT test explained that there was no valid result. Skin Prick Test for patients suffering from allergic rhinitis showed 95% of respondents experience of positive result and the other 5% did not. This was possible because allergic rhinitis diagnosis is based on clinical and physical test with a high level of eosinophil and or IgE serum; hence it triggered negative result on SPT test. Skin prick technic, allergen type, and pricking affect the result of SPT. This study uses disposable synthetic lancet from Stallergenes Lancet/ Stallerpoint® to minimize different result because of skin prick or the medical staff.[18]

The cause of negative allergic rhinitis patient were explained by different allergen extract used in testing process and original allergen inside patients' body; thus the sensitivity was different and gave a negative result. Another possible cause was a fact that patients practice daily diet towards allergen trigger before receiving treatment from the hospital. Thus, the specific IgE level inside the blood was too low and did not cause any reaction. Splitting time of IgE that needed only 2.5 days could explain about this symptom.[14] Another possibility was allergen type of the patients which could not match with allergen extract used in experiment; so there was no reaction found in SPT.

SPT showed with various results. The most frequent allergen type in SPT is was *Dermatophagoides pteronyssinus* (79%), *Dermatophagoides farinae* (72%) and cockroach (70%). It is aligned with a previous research conducted in Medan stating that most allergens found based on SPT are dust mites (43.5%) and cockroach (41.9%).[19] This result is contradictive towards one conducted in Malaysia stating that most allergens was *D.farinae* (79.69%) and *D.pteronyssinus* (68.42%).[5]

The amount of positive result of non-patients with allergic and non-infectious rhinitis comparative group taking SPT were six persons (35%) with Inhalant allergen was *D.farinae* 4 (23.53%), *Alternaria* 4 (23.53%), *D.pteronyssinus* 3 (17.65%), *Aspergillus* 3 (17.65%), cockroach 3 (17.65%) and *cat fur* three (3) (17.65%). Ingested allergens mostly caused by tuna fish 4 (23.53%), peanut 3 (17.65%), eggs yolk 2 (11.7%), crab 2 (11.7%), shrimp 2 (11.7%) and wheat flour 2 (11.7%).

Positive result of comparative group was acquired because there was group separation before SPT procedure; thus there were positive results amongst the expected negatives. False positive outcomes occurred in the group of patients not suffering from allergic and non-infectious rhinitis could be caused by high level of skin's sensitivity as the aftermath of trauma in prick test session. Then, irritation and dermographism, allergen extract contamination and excessive reaction could not be avoided as well.[20] Other factors that may trigger are clinical symptoms such as: history of allergic records and physical test with or without eosinophil in peripheral blood and or the amount of IgE serum in diagnosing rhinitis, thus it may generate false outcomes.

The positive results from specific IgE test based on *Foresight*® immunoblot methodology to the froup of patients with allergic rhinitis were counted in 36 people (84%) with the most Inhalant allergen: *D.farinae* and *D.pteonyssinus* (D1/D2) in 29 people (67.44%). The research in Korea using immunoblot method from *AdvanSure Allergysscreen* stated that the most allergen obtained are *D.farinae* followed by *D.pteronyssinus*.[5,21] The cause of negative result on specific IgE test based on *Foresight*® immunoblot methodology in patients with allergic rhinitis were: the difference allergen extract blotted in immunoblot line with the origins of allergen inside the patients' body, thus, its sensitivity is different and it gives negative outcomes. Besides, diet life style towards the cause of allergen could decrease the level of specific IgE and it fails to bind with antigen-antibody in immunoblot line. It could be explained by half time of IgE that is only 2.5 days.[14,22] Another possibility is allergen type inside patients' body other than allergen extract blotted in immunoblot lines, *Foresight*® in this research, hence, it would give positive outcomes. The false negative result could be caused by invalid reagent and wrong procedures from *Acon Laboratories* and using well-package and stored reagent.

The positive outcomes from specific IgE test based on *Foresight*® immunoblot methodology in control group of patients with rhinitis not caused by allergic and non-infectious disease is 5 people (29%) with Inhalant allergen; ranged from *D.farinae* and *D.pteonyssinus* (D1/D2) summed in 6 people (14%). In Ingestan allergen test, there was no positive outcome.

The specific IgE test based on *Foresight*® immunoblot methodology on control group of patients

with rhinitis not caused by allergy and non-infection in this research shows 29% positives and 71% negatives. This study enabled positive outcomes in control group (false positive result) because of group separation was only based on clinical signs, history of allergic medical records, physical test, or without the IgE serum level and or blood eosinophil so the false positive outcomes appeared. It was also caused by contamination of serum samples and unsufficient membrane moistened treatment.[6]

The allergen pattern from 16 SPT showed possibility patients with allergic rhinitis having more than one allergen. Each allergen could appear by itself or simultaneoulsy. There was no patient who had single allergen. It was in line with a previous study conducted in Medan results on multiple studies in 2007.[19] Multiple allergens result in SPT based on studies in Turkey, 2012, which stated that there was no significant relation between fatality of disease and multiple SPT.[23]

The result on IgE specific test based on *Foresight*® immunoblot methodology enabled patient to be diagnosed as having more than one allergen. Each allergen would appear by itself or simultaneously. Allergic patient, generally, had a high level of sensitivity towards multi-allergens. Thus, it caused difficulty to determine main and important allergen.[10] Most allergens in this study could be used as a perimeter to allergic rhinitis treatment and prevention. Figure 3 shows that the dissemination pattern of allergen is mostly from testing specific IgE based on *Foresight*® immunoblot methodology.

D1/D2: *D.pteronyssinus, D.farinae*; E1/E5: cat fur, dog fur; I6: cockroach; F4: wheat flour; F1: egg's yolk; F3/F24/F23: fish, shrimp, crab; W1/W6: *Ragweed, mugwort*; F202/F13/F14: cashew peanut, ground nut, soybean; M1/M2/M3/M6: *Alternaria alternate*, *Aspergillus fumigatus*, *Penicillium chrysogenum*, *Cladosporium herbarum*

The test of specific IgE through *Foresight*® immunoblot methodology is a semi-quantitative laboratory experiment; hence it is difficult to determine the exact value of it. It also has high detectability; however the minimum saturation of specific IgE that could be detected is approximately at 0.35 IU/mL.

The result of testing specific IgE based on *Foresight*® immunoblot methodology shows that patients with positive allergic rhinitis are 36 (83.7%)

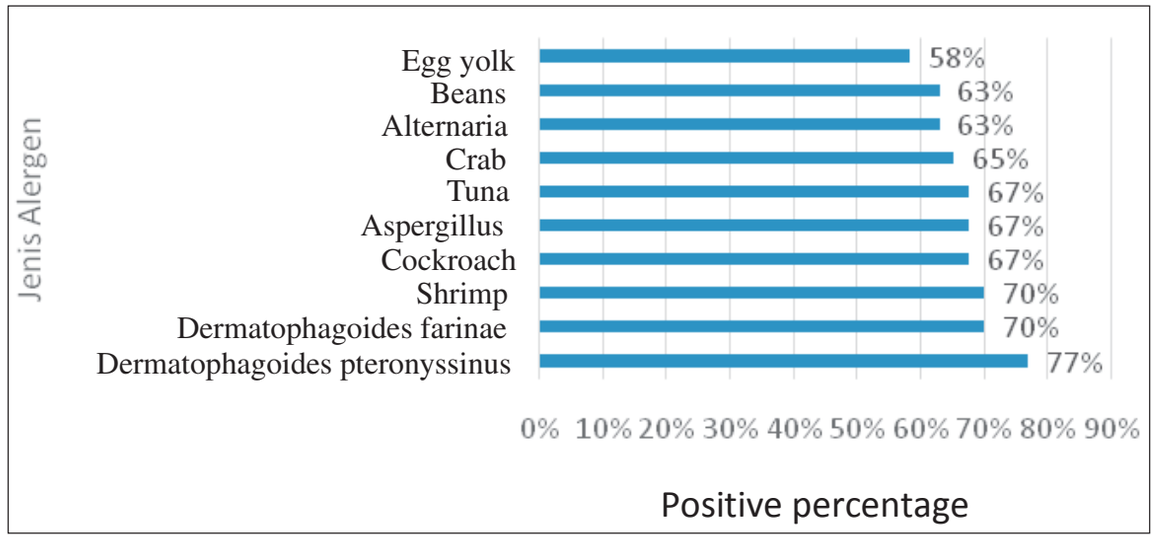

Figure 2. Allergen pattern of allergic rhinitis with SPT

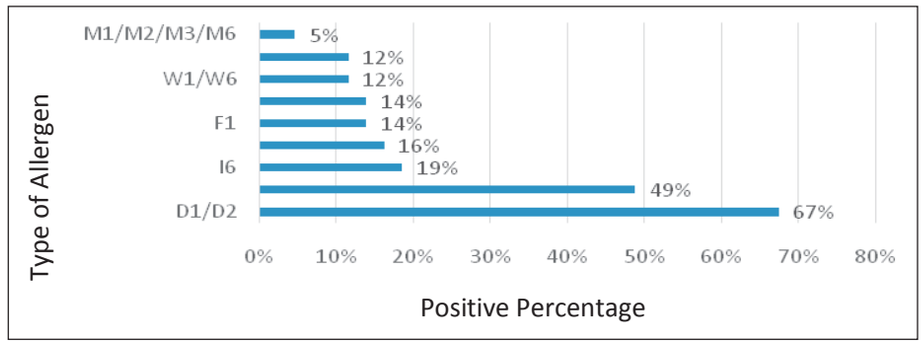

Figure 3. Allergen pattern of allergic rhinitis patients based on IgE experiment by *Foresight*® immunoblot methodology

Tabel 2. The result of testing specific IgE based on *Foresight*® immunoblot methodology and SPT to patients with allergic rhinitis and non-allergic and non-infectious rhinitis

Diagnosis	*Skin Prick Test*				IgE based on *Foresight*® immunoblot methodology			
	Postive		Negative		Positive		Negative	
	N	%	N	%	N	%	N	%
Rhinitis caused by allergy	41	95,3	2	4,7	36	83,7	7	16,3
Non-allergic and non-infectious rhinitis	6	35,3	11	64,7	5	29,4	12	70,6

Table 3. The diagnostic value of specific IgE testing based on *Foresight*® immunoblot methodology towards allergic rhinitis detected by Skin Prick Test

Statistical analysis	Specific IgE based on *Foresight*® Immunoblot methodology	Range values (confidence interval 95%)
Diagnostic sensitivity (%)	72.34	58.24–83.06
Diagnostic specificity (%)	46.15	23.21–70.86
Positive predictive value (%)	82.93	68.74–91.47
Negative predictive value (%)	31.58	15.36–53.99
Diagnostic feasibility (%)	66.67	54.06–77.27
Positive probability ratio	1.343	0.9932–1.817
Negative probability ratio	0.5993	0.3521–1.02

and negative allergic rhinitis were 7 (16.3%); however result for patients with non-allergic rhinitis and non-infection showed that negative outcomes in 5 persons (29.4%) and positive outcomes in 12 persons (70.6%).

The outcomes of Fisher analysis was p=0.312. Diagnostic test value specific IgE based on *Foresight*® towards SPT is demonstrated in Table 3. The sensitivity of diagnosis based on immunoblot methodology towards SPT was 72.34% (confidence interval: 95%; 58.24–83.06) in which was categorized as high level. The feasibility of specific IgE testing based on immunoblot methodology compared to SPT in diagnosing allergic rhinitis accurately was 72.34% and the other 27.66% of patients were lost from the diagnosis (semi negative). Its sensitivity was affected by multiple allergen layered in immunoblot line; thus it filters as much as possible. Line or allergen tape inside the immunoblot containing several allergens had similar protein structure; hence it could easly catch more specific IgE.[6]

Anoher study done in Korea[21] stated that the sensitivity of other MAST-Immunoblot towards SPT was between 27–63% which means to have lower sensitivity compared to *Foresight*® immunoblot. The difference was possibly because of cut off differences on specific IgE level and used allergen.

The particular diagnosis on specific IgE testing based on immunoblot methodology towards SPT in general, in this study, showed at 46.15% (margin confidence interval: 95%; 23.21–70.86). This condition could be said as having lower unique characteristics. It was affected by many things; excessive antibody saturation, unspecific and unexpected bonding places inside the membrane, antibody complex and matrix effect attached to nitrocellulose used on it.[24]

It was contradictive with the previous study held in Korea towards the uniqueness of AS immunoblot compared to SPT in which was found between 81-97%.[21] The research conducted in 2007 showed that the result of specific and different IgE reagents it could not be compared to one another even though they are presented in similar unit.[25] The sensitivity of diagnostic of specific IgE testing based on immunoblot methodology towards SPT was higher than its diagnostic uniqueness, 72.34% and 46.15%. It was suitable with disease filtering necessity and it could not be determined as allergen before curing it with specific immune. It was not compatible with the previous study conducted in Malaysia based on other immunoblot methodology with sensitivity in 63.16% and uniqueness in 65.57%.[5] Defining allergens before specific immune injection could not be executed simultaneously with specific IgE testing based on immunoblot methodology. It was because in one of immunoblot line contained more than one allergen.

In this research, positive estimation value was obtained at 82.93% (confidence interval: 95%; 68.74–94.47). It clearly stated from 43 subjects; there were 34 people acquiring positive results on specific IgE testing based on immunoblot methodology. Thus, it meant that positive estimation value was categorized as high level; in the other words, the feasibility of such test was good to determine allergic rhinitis. The cause of pseudo-positive result could be found amongst the infectious disease from parasite, autoimmune disorder and other diseases caused from immunodeficiencies

Table 4. The comparison between diagnostic value of SPT and specific IgE based on *Foresight*® immunoblot methodology towards allergic rhinitis diagnosis based on battery test

Statistical analysis towards patients' diagnosis	SPT	Specific IgE based on *Foresight*® Immunoblot methodology
Diagnostic sensitivity (%)	90.3	87.1
Diagnostic specificity (%)	34.5	51.7
Positive predictive value (%)	59.6	65.9
Negative predictive value (%)	76.9	78.9
Positive probability ratio	1.379	1.804
Negative probability ratio	3.563	4.009
Kappa coefficient	0.253	0.393
P	0.020	0.001

such as Wiskott-Aldrich and DiGeorge syndrome and hyperimmunoglobulin E.[14,26]

Negative predictive value of specific IgE testing based on immunoblot methodology was 31.58% (confidence interval: 95%; 15.36–53.99). It showed that 6 out of 17 subjects really obtain negative result on the test. The value was categorized as medium level. It portrays that negative functionality of specific IgE based on such methodology to determine allergen within patients' body was entirely good.

The feasibility of diagnosis of specific IgE testing based on immunoblot methodology was 66.67% (confidence interval 66.67; 54.06–77.27). This feasibility was composed of sensitivity and specificity from particular laboratory experiment. Result of feasibility level of diagnosis proved that the function of specific IgE test based on immunoblot methodology to determine allergic rhinitis diagnosis was convincing. If there was any possibility to diagnosis misconduct, it would be only 33.33%. Positive probability ratio estimation is 1.343 which means specific IgE testing based on *Foresight*® immunoblot methodology has 1.343 points on allergic rhinitis diagnosis compared to non-allergic and non-infectious rhinitis. Negative probability ratio estimation was 0.5993 which means specific IgE testing based on immunoblot methodology was not possible to be determined as allergic rhinitis diagnosis at 0.5993 compared to non-allergic and non-infectious rhinitis. The data of diagnostic value of specific IgE based on immunoblot methodology stated that it had good validity.

The aim of this research was to know conformity level of specific IgE diagnosis based on *Foresight*® immunoblot methodology with SPT and using a battery test on allergic rhinitis as the main factor (check up: clinical test, history, the entire IgE serum and eosinophil in peripheral blood) (Table 4). By determining it declared that the actually hypothesis was rejected because there was no conformity between specific IgE testing based on *Foresight*® immunoblot methodology and SPT.

If the diagnostic value of SPT and specific IgE immunoblot was compared to allergy battery test as gold standard; it would show result with higher kappa coefficient of specific IgE than SPT, statistically speaking. Positive predictive value of specific IgE based on *Foresight*® immunoblot methodology or the feasibility of positive specific IgE based on immunoblot methodology to detect allergic rhinitis was higher than SPT. Negative predictive value showed that specific IgE level was higher than SPT. Specific Immunoglobulin E had better quality in terms of its specificity so it helped patients to determine allergic rhinitis.

The result of specific IgE based on *Foresight*® immunoblot methodology between patients with allergic rhinitis and non-allergic also non-infectious rhinitis was processed by Mann-Whitney test and showed p=0.000 (confidence interval=95%). Its significant difference between two groups was at p <0.05. The procedure could be used to differentiate allergic rhinitis and non-allergic as well as non-infectious rhinitis even though there was no or only a little conformity with SPT. If the result shows positive result from healthy people, thus it should be followed by clinical check up to determine further treatment.[21]

CONCLUSION AND SUGGESTIONS

In conclusion, most allergens amongst patients with allergic rhinitis based on immunoblot result in the Dr. Soetomo Hospital Surabaya were *D.farinae* and *D.pteonyssinus* (D1/D2), around 29 (67.44%); specific IgE testing based on immunoblot methodology compared to SPT had following diagnostic values: sensitivity 72.34%, specificity 46.15% and feasibility 66.67%, positive predictive value 82,93% and negative value 31.58%, positive probability ratio 1.343 and negative probability ratio was 0.5993. This result showed there was no conformity between specific IgE testing based on immunoblot methodology and SPT; because each had different diagnostic value so that

it could be used simultaneously to diagnose and cure allergic rhinitis.

The recommendation for further research is as follows: the study needs to avoid pra-analysis contaminant (diet of rhinitis patients, consumption and ceasing of steroid medicine and antihistamine) and analysis contaminant from specific IgE testing based on immunoblot methodology (using appropriate properties: stock of immunoblot reader and flexible shaker); additional panel test to detect allergy and infection (eosinophil dredging-nose and CRP); do further research with patients experiencing urticaria or active eczema who are impossible to receive SPT; thus, the advantages of specific IgE testing based on immunoblot methodology is revealed.

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