

Estimation of Specific Immunoglobulins E,A Antibodies and E-rosette in Allergic Patients.

Ishraq Abdul Amer Saleh ALmaamory
Science For Women / University of Babylon
mali:asho2091@yahoo.com.

Abstract:

Background: Allergy is a hypersensitivity reaction mediated by immunological mechanisms which can be antibody or cell-mediated.

Objective: This study aims at evaluating serum immunoglobulins A and E levels in patients suffering from Allergic in comparison with healthy individuals.

Methods: This work was applied on 87 Allergic patients admitted to the Babylon Center of Allergic and 40 apparently health controls with age range (10-69 years).

Results: The Immunological parameters showing that there is a significantly increased ($p < 0.05$) in IgE(448.75) IU/ml compared to control group (52.11) in age group 10-19 and a significantly increased ($p < 0.05$) in IgA(380.5) mg/ml compared to control group (110.54) in age group 50-59 and no significantly increased ($p > 0.05$) of E-rosette test level in the allergic patients (32.601%) compared to control group (17.224%) in age group 50-59 years.

Conclusion: We conclude that there is an association of all Allergic IgE level increase it in differant age also IgA level in serum of Allergic.

Key words: Allergic , IgE Elisa , IgA Elisa, E-rosette test.

الخلاصة:

أعدت دراسة مناعية لمعرفة مدى شيوع الأجسام المضادة IgE, IgA كذلك اجري اختبار E-rosette لمصول مرضى الحساسية في محافظة بابل اللذين كان عددهم 87 مريض و للفئات العمرية بين (10-69) سنة مقارنة بالاصحاء اللذين بلغ عددهم 40 شخص لكلا الجنسين .

تم تقدير نسبة IgE و IgA للمرضى والأسوياء باستعمال تقنية الامتزاز المناعي المرتبط بالأنزيم (ELISA)، أظهرت الدراسة أن هنالك فارق معنوي ($P > 0.05$) بين مصول مرضى الحساسية مقارنة بالسيطرة وجد أعلى تركيز IgE بلغ 448,75 وحدة دولية /مل مقارنة بالاصحاء اللذين بلغ تركيز مصولهم 52.11 عند الفئة العمرية 10-19 وفي حين بلغ تركيز IgA 380.5 ملغم /مل مقارنة بالاصحاء اللذين بلغ تركيزهم 110.54 ملغم/مل عند الفئة العمرية 50-59 سنة IgA في حين بلغت نسبة E-rosette للمرضى 32.601% مقارنة بالاصحاء التي بلغت نسبتهم 17.224% عند الفئة العمرية (50-59) سنة .

كما بينت الدراسة الحالية وجود ارتفاع في معدل تركيز الأضداد IgE و IgA في مصول المرضى الحساسية بالمقارنة مع مجموعة السيطرة .

الكلمات المفتاحية : الحساسية ، IgE Elisa , IgA Elisa, E-rosette test.

Introduction

Allergic is characterized by acute episodes of airway obstruction precipitated by respiratory infection and the release of IgE dependent mediators ,airway inflammation resulting from an inappropriate response to either infectious or allergic antigens is a finding common to the different manifestations of asthma . (Ozcan *et al.*,2008)

An allergic reaction is caused when a person's immune system produces IgE antibodies in response to a foreign antigen (allergen). IgE molecules are tightly bound to the surface of mast cells and basophiles in blood. These cells contain granules that have a high concentration of histamine and other substances that are responsible for the allergic reaction (Vercelli,2000).

Upon subsequent exposure to the same antigen,an immediate(type1) hypersensitivity reaction called the atopic or allergic reaction ensues.The allergen binds to the IgE and the cross linking of antigen and antibody molecules causes the mast cell to degranulate. The histamine and other allergic mediators are released and cause local swelling(edema) and redness

(vasodilation). These reactions occur immediately and may be sufficient in intensity to cause constriction of the bronchi and shock.

Such asystemic response to an allergen is called an anaphylactic reaction. Allergens most often responsible for anaphylactic reactions are insect bites and penicillin in persons who are allergic to these agents.(Melvin and Ramanathan 2012).

A selective deficiency, defined by serum IgA of less than 0.05mg is the most frequent human defect in humoral immunity,with a prevalence of 1/2,000 to 1/500. This mechanisms of class switching to IgA defect, affecting mucosal and systemic plasma cells, is due to genetic defects .(Phillips *et al.*,2001).The healthy, selective IgA deficiency has been associated with an increased prevalence of atopy, food sensitization, recurrent infections, and neoplastic and autoimmune disorders. (Peebles *et al.*,2001)

Materials and Methods

Patients and Control

A total of 87 Allergic patients consisting of 40 healthy controls were involved in this study,their age range was from (10–69) years.Case information was taken for each patient include; name, sex, age, residency, duration of infection and duration of therapy.

All Allergic cases were clinically diagnosed by a specialist clinician.Those patients were admitted to the Babylon Center of Allergic .

Atotal of 40 apparently healthy subjects were involved as controls group. The age range of controls was matched to the patients (10– 69) years.

Blood samples:

Three ml of blood were collected by vein puncture into two sterile test tubes , in one of them 2 ml of blood were put and left for (2–4) hours,then the upper layer (serum)was collected in clean test tube and stored at–20 C° until using it in serological tests and determination of IgE,IgA .The second sample of blood 1 ml was put in another test tube containing anticoagulant EDTA to E-rosette test (Lewis *et al.*,2001).

Estimation the concentration of IgE,IgA .

In vitro test which used enzyme –linked immunosorbent technology which measures, bacteria antigens number or enzyme- linked immunosorbent assay (ELISA) technology IgE,IgA production in whole blood (Lazarevic *et al.*,2005).

Cellular immune function tests:

E-rosette tests (T-cell count):

Based on the fact that the T-lymphocytes were considered E⁺-rosette forming T-cells when three or more sheep erythrocytes adhered to it, otherwise; they were regarded as E⁻-rosette forming T-cells.

Statistical Analysis

T-test (p <0.05) were carried out according to (Niazi,2004).

Results and Discussion

Age Distribution:

Many Allergy cases were diagnosed in 10-19 years old young child, the number increased in the age group of 50-59 years and also increased allergy case in 50-59 years, table (2). However, the number increased in the age group of 10-19 to reach 22% of the total number of Allergy patients.The number of cases were maintained in most at the same level in the age groups of 50-59 years (20.6%) and 40-49 years (18 %).Only 10.3% with the 60-69 years age group. The results that were expressed in table (1) revealed the wide age range for Allergy patients. In child allergy is high than adult because immune system in small age less than in order and the reaction allergic can come in different ways rashes, fever ,breathing

problems but if can also only be mind symptoms, such as a runny nose, sore throat or an unusual tiredness.

(Rondon *et al.*,2010)

Table (1) Age Distribution for Allergic Patients.

Age group (years)	Allergic patients
10-19	20: 87 (22 %)
20-29	10: 87 (11.4 %)
30-39	14: 87 (16.09 %)
40-49	16: 87 (18 %)
50-59	18: 87 (20.6%)
60-69	9:87 (10.3%)
Age range (years)	10-69

Sex Distribution:

In this study, the Allergic patients consisted of 62:87 (71.2%) males and 25:87 (28.7%) females, figure (1). For patients showed that male–female ratio was higher in males than in females. This finding was matched with that recorded by (Kim *et al.*,2011) who mentioned that the rate of Allergic in male was higher than female for Allergic patients attending the Babylon Center of Allergic . This difference is partly due to the fact that man have less access to diagnostic facilities in some settings, but the broader pattern also reflects real epidemiological differences between men and women, both in exposure to infection and in susceptibility to disease.(Szakos *et al.*,2004) .

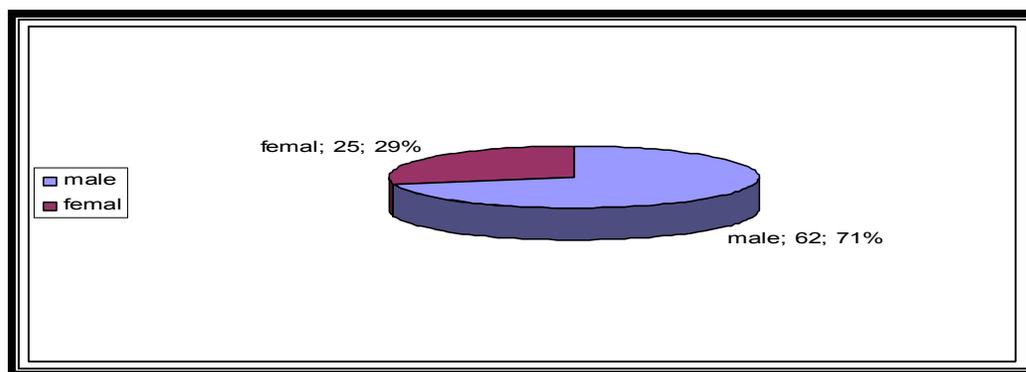


Figure (1) Sex Distribution for Allergic Patients.

Concentration of IgE.

In this study, it was observed that IgE levels in population were higher than control. The allergic patients have an expected in age group 10-19 IgE concentration up to 448.75 IU/ml while in control IgE levels concentration appears 50.11 IU/ml significantly($p < 0.005$) (Table 2) .The higher IgE levels in allergic explained probably by the higher incidence of parasitic infestations and allergic complication (Gennaro *et al.*,2011). The biological activities of IgE in allergic patients, had been reported by Rondon and his colleagues (Galli *et al.*,2008) The biological role of IgE is inducing humoral and cellular immune responses, increased levels of IgE and inflammatory cells, are related with Th1 and Th2 responses in patients with allergic (Kaneko *et al.*,2012).

Table (2) Concentration of IgE IU/ml for Allergic Patients and Controls

Age group (years)	Group	IgE IU/ml
10-19	Patient	*448.75±33.10
	Control	50.11±9.09
20-29	Patient	* 227.42± 18.9
	Control	78.79±9.85
30-39	Patient	*431.14±27.15
	Control	83.68±3.56
40-49	Patient	*228.58±28.9
	Control	58.48±1.73
50-59	Patient	*346.19±37.18
	Control	75.00±3.50
60-69	Patient	*221.52±25.61
	Control	74.48±3.008

***Standard deviation**

The allergic patients have an expected IgA concentration up to 380.5mg/dl while in control IgA levels concentration appears 110.54 mg/dl in age group 50-59 significantly ($p < 0.005$) The result shows in (Table 3) .

It may be stimulating allergen-specific B-cells in the bronchial mucosa to produce specific IgA antibodies.(Fouda,2004)The nature of serum IgA activation in response to the different factors in allergic patients remains to be elucidated.

In conclusion, the association levels of IgE and IgA with allergic patients were investigated. The results revealed that mean serum IgE levels were significantly higher in patients than controls. We also found that mean serum IgA levels were significant different in allergic patients compared to controls which may have predictive roles in the allergic patients, the IgA plays a role in host defense against infection.(Oftedal *et al.*,2007)

Table (3) Concentration of IgA levels (mg/dl) for Allergic Patients and Controls.

Age group (years)	Group	IgA mg/dl
10-19	Patient	181.5 ±52.8
	Control	80.54 ±30.2
20-29	Patient	201.5 ±42.74
	Control	78.44±25.2
30-39	Patient	330.5 ±72.74
	Control	88.22±12.2
40-49	Patient	234.5 ±72.74
	Control	75.22±19.4
50-59	Patient	380.5 ±33.74
	Control	110.54 ±30.2
60-69	Patient	190.5 ±62.74
	Control	95.22±18.4

*Standard deviation

**The cellular immune function tests:
E-rosette Formation (T-cells count)**

E-rosette is a simplified common technique used for the quantitative assay of T-lymphocytes. It is different from EAC -rosette test, which is used as a tool for the quantitative assay of B-lymphocytes. E -rosette test needs for the interaction of antibodies (A), complement (C), sheep erythrocytes (E) and specific ligands on the surface of B-lymphocytes, while in the case of T-lymphocytes, the interaction occurs directly between sheep erythrocytes and specific T-cell receptors (CD₂) (13.14). In addition to CD₂, (15) reported that another T-cell surface molecules (32kDa, 20kDa and E2) may be involved in spontaneous rosette formation with sheep RBCs.

The result table (4) showed that there were gradual patterns for the differences in the values of E-rosette test. Generally, the values of Allergic were higher than control. The immunological anergic state is induced in allergic patients when the CD₄ T lymphocytes count is sub normal (Quercia *et al.*,2012).

Table (4): E-rosette test for Allergic and control Patients.

Age group (years)	Group	E-rosette
10-19	Patient	*28.347±4.646
	Control	16.295±2.1836
20-29	Patient	27.666±6.371
	Control	14.345±0.678
30-39	Patient	28.662±7.168
	Control	13.913±1.662
40-49	Patient	30.463±6.246
	Control	13.284±1.032
50-59	Patient	32.601±3.22
	Control	17.224±1.921
60-69	Patient	28.123±2.21
	Control	15.524±2.33

References

- Fouda EE. 2004 Immunoglobulin A (IgA) in allergic airway disease. The Journal of Allergy and Clinical Immunology. ;113:328
- Galli SJ, Tsai M and Piliponsky AM. 2008 . The development of allergic inflammation. Nature. ;454: 445–454.
- Gennaro D'Amato, Chair, MD, Menachem and Rottem, MD, 2011 , and for the WAO Special Committee on Climate Change and Allergy Climate change, migration and allergic respiratory diseases: An update for the allergist. World Allergy Org J.; 4 (7): 120-125
- Kaneko M, Jarjour N, Swanson M, Busse W, Gleich J and Kita H. 2012). Allergen-specific IgG and IgA in broncho alveolar lavage fluids from patients with allergy induce eosinophil degranulation. J Allergy Clin Immunol ;95: 339-142.
- Kim, D.; Sato, A.; Fukuyama, S. and Sagara, H. 2011 .The airway antigen sampling system : Respiratory m cells as an alternative gateway for inhaled antigens . Lippincott Williams Wilkins, London , PP. 87 - 104.
- Lazarevic, V.; Pawar, S. and Flynn, J. 2005 . Measuring T-cell function in animal models of asthmatic by Elispot Methods. Mol. Biol. 302: 179-190
- Lewis, S.M.; Bain, B.J., and Bates, I. 2001 . Practical Haematology. 9th ed. Churchill Livingstone, London.
- Melvin TA and Ramanathan M Jr. 2012 Role of innate immunity in the pathogenesis of allergic rhinitis. Curr Opin Otolaryngol Head Neck Surg. 20:194-198.
- Niazi, A. 2004 Statistical analysis in medical research .2nded .College of Medicine , Nahrain University. Baghdad .PP. 73-98.
- Oftedal B, Brunekreef B, Nystad W and Nafstad P. 2007 . Residential outdoor air pollution and allergen sensitization in school children in Oslo, Norway. Clin Exp Allergy. 37:1632–1640

- Ozcan E, Notarangelo LD and Geha RS 2008 . Primary immune deficiencies with aberrant IgE production. *J. Allergy Clin. Immunol.*122:1054.
- Peebles RS Jr, Hamilton RG, Lichtenstein LM, Schlosberg M, LiuMC, Proud D and Togias A. 2001 . Antigen-specific IgE and IgA antibodies in broncho alveolar lavage fluid are associated with stronger antigen induced late phase reactions. *Clin Exp Allergy* ;31:239–248.
- Phillips JO, Everson MP, Moldoveanu Z, Lue C and, Mestecky M. 2001 . Effect of IL-4 and IFN-gamma on the expression of polymeric Ig receptor (secretory component) and IgA binding by human epithelial cells. *J Immunol* ;145:1740–1744.
- Quercia O, Incorvaia C, Puccinelli P, Scurati S, Emiliani F and Frati F, 2012 . Prevalence of allergic disorders in Italy: the Cotignola population study. *Eur Ann Allergy Clin Immunol.*; 44:5-11.
- Rondon C, Canto G and Blanca M 2010 . Local allergic rhinitis: a new entity, characterization and further studies. *Curr Opin Allergy Clin Immunol.*; 10:1-7
- Szakos, E., Lakos, G. and, Aleksza, M. 2004 . Association between the occurrence of anticardiolipin IgM and mite allergen-specific IgE antibodies in children with extrinsic type of atopic eczema/ dermatitis syndrome. *Allergy*,59(2):164-167.
- Vercelli D 2005. Genetic regulation of IgE responses: Achilles and the tortoise. *J. Allergy Clin. Immunol.* 116:60.