

Immunological aspects on Asthmatic Patients

Dr.Mayada Farhan Darwesh

Collage of Science, kufa university

Abstract

The study was designe to assess some immunological aspects of asthma patients by detected the total and differential count of leucocyte , immunoglobulin titer and specific activity of adenosine deaminase ADA . Therty sex samples of patients were collected and 24 sample of healthy as control group .

The result was revealed that the WBCs count increased in patients group compared with control group , and the neutrophils and basophile were increased significantly while, lymphocyte and monocyte was decreased.

The concentration of IgE increased significantly but IgG was decreased
The specific activity of ADA was **highly significant ($P \leq 0.01$) increased** compared with control group.

Introduction

Asthma is a common chronic disorder of the airways that involves a complex interaction of airflow obstruction, bronchial hyperresponsiveness and an underlying inflammation. This interaction can be highly variable among patients and within patients over time. The interaction of these features of asthma determines the clinical manifestations and severity of asthma and the response to treatment (Busse and Lemanske., 2001). Airflow limitation in asthma is recurrent and caused by a variety of changes in the airway. These include: Bronchoconstriction, Airway edema, Airway hyperresponsiveness , Airway remodeling, (Holgate and Polosa 2006).

In asthma and other allergic diseases, the immune system senses for allergens, perceives them as foreign, and begins to prepare to fight off them off as a foreign intruder. The process that takes place is often referred to as the allergic cascade, which generally occurs in 3 steps:

1-Sensitization- Allergen Exposure , 2- Early Phase Response- Re-exposure, 3- Late Phase Response, The first time which exposed to an allergen, that stimulate the allergic cascade through: Inhalation of substances such as dander, pollen or dust mites, or Ingestion of foods or medicines, or Physical contact of skin with substances such as poison ivy. Immunologically, the body senses the allergen as foreign and sets off a cascade of events stimulating several different types of immune cells : T cells rapidly stimulate B cells (which transform into plasma cells then produce IgE antibodies specific to the allergen) ,IgE antibodies bind to mast cells . During subsequent exposures to the allergen, may develop asthma symptoms as part of the early-phase response. With re-exposure to the allergen immune system senses the allergen as foreign leading to: The mast cell/ IgE complexes produced in the sensitization phase, binding to the allergen thinking that it is a foreign invader, then mast cells release inflammatory cells called mediators (e.g. histamine) that quickly travel throughout body with the purpose of fighting off the foreign invaders such as bacteria and viruses. This mediators cause allergy symptoms(wheezing, coughing or feeling short of breath as the immunologic response causes swelling and narrowing of the airways in lung, during late phase response. Mediators released by the re-exposure to an allergen also stimulate other kinds of immune cells called eosinophils.

Eosinophils contain substances that when released normally fight off infections, but in asthma, the cells damage the lung causing more inflammation and worsening symptoms (Fahy *et al.*,1995 ; Jatakanon *et al.*,1999) .

Adenosine is a potent and ubiquitous signaling molecule, which accumulates in times of cellular stress and damage. At sites of inflammation, adenosine is generated by the catabolic breakdown of ATP and functions to regulate the inflammatory response through engagement of putative adenosine receptors. Consistent with this, asthmatic patients have increased adenosine levels in their BAL fluid and exhaled breath condensates, and the degree of adenosine elevations correlate with disease severity (Huszar *et al.*,2003) . These findings suggest that adenosine may be involved in the disease pathogenesis of chronically inflamed asthmatic lungs, including angiogenesis.

Adenosine deaminase is polymorphic enzyme found in most human tissue catalysis the hydrolytic deamination of adenosine and deoxyadenosine to inosine and deoxyinosine respectively(Casoli,*et al.*,1988). Clinical interest in this enzyme has been revived by the discovery of asyndrome of severe humoral and cellular immunodeficiency associated with deficiency of ADA in erythrocytes (Giblett *et al.*,1972) .

The present study was aimed to determined some immunological parameters in asthmatic patients which include :

- 1- *White blood cells count and deferential count*
- 2- *Immunoglobulin titer*
- 3- *Specific activity of adenosine deaminase determination .*

Materials and Methods

* Study groups: 36 patients admitted to the a sensitivity and asthma center in the AL-Sadder hospital. These patients were different ages and they suffer from asthma . In addition to 24 healthy persons different ages have been treated as controls .

* Blood samples: peripheral blood was withdrawn from patients by sterile syringe usually in the range of (5-10 ml) . The blood was transferred into two tubes under aseptic conditions , one for white blood count and differential count (sterile heparinized tubes) and the another was used for serum collection . Blood samples from healthy were assayed identically .

* White blood cells count and differential count: were carried in the symix apparatus (AL-Sadder hospital)

* Signal radial diffusion test :procedure based on Mancini *et al.*,(1965)

* Enzymatic activity : The activity of adenosine deaminase (ADA) was determined in serum according to Giuist (1981) .

* Protein determination :The protein concentration in the serum was determent according to Bradford.,(1976) .

Results and Discussion

The results in the Table (1) show that the significant increase in WBCs were count compared with control group , the neutrophile, Basophile cells were highly significant ($P \leq 0.01$) increased while the monocyte and lymphocyte cells were decreased in compared with control group .

Table 1 : White blood count and diffrenial count in asthmatic patient compared with control .

	WBCs count Mean \pm S.E	Lymphocyte Mean \pm S.E	Neutrophile Mean \pm S.E	Basophile Mean \pm S.E	Monocyte Mean \pm S.E
Control	8400 \pm 2000	54 \pm 13	25 \pm 9	9 \pm 2	10 \pm 0.5
Asthma patient	12.300 \pm 2800	40 \pm 11	31 \pm 10	20 \pm 4	6 \pm 0.3

Histamine is the inflammatory compound released during allergic reactions that causes runny nose, watery eyes, and wheezing, it could be produced in large amounts in the lung by neutrophils, the white blood cells that are the major component of pus (Galli *et al.* ,2005) . Neutrophils are increased in the airways and sputum of persons who have severe asthma, during acute exacerbations, and smoking (Fahy *et al.* 1995). Onther study explain that the primary sources of lung histamine, in health as well as patients, are mast cells, which are classically associated with allergy (Caughey *et al.*,2007) .The results could mean that histamine acts as a link between airway infections and asthma and bronchitis, which are associated with allergy . Basophils,also containing histamine which are involved in some allergic reactions.

- The immunoglobuline titer in sera of the patients were studied to compared with control group. Results were presented in Table (2) show that IgG concentration are significantly reduced in asthmatics but IgE concentration was significantly increased , IgM concecntration was no change compared with control group.

Table 2 :Immunoglobulin titer in asthmatic patient compared with control group .

	IgG	IgM	IgE	IgA
Control	851.21 \pm 32.11	125.80 \pm 23.66	85 \pm 20.21	97.96 \pm 39.25
Asthma patient	677.45 \pm 45.62	130.74 \pm 43.55	101 \pm 35.11	77.29 \pm 28.67

IgE is the antibody responsible for activation of allergic reactions and is important to the pathogenesis of allergic diseases and the development and persistence of inflammation. IgE attaches to cell surfaces via a specific high-affinity receptor. The mast cell has large numbers of IgE receptors ;when activated by interaction with antigen, release a wide

variety of mediators to initiate acute bronchospasm and also to release pro-inflammatory cytokines to perpetuate underlying airway inflammation (Boyce 2003) . Holgate *et al.*, (2005) shown that the reduction of IgE is effective in asthma treatment these results agree with previous studies published by (Louftus *et al.*,1988), which explain that defects of the immune system that affect exclusion or elimination of antigen could be relevant in early infancy at the time of first presentation of antigen to cells making IgE, clearance of adsorbed antigen might be impaired in IgG deficiency . Inability to mount an adequate IgG response might provoke or exacerbate asthma in certain children. Respiratory infection, particularly when viral, results in an increase in bronchial reactivity. (Empey .,1976) Perhaps protracted infection and epithelial damage enhances this effect on airway function . Extrinsic asthma is an allergic condition with an increased level of IgE antibodies due to environmental causes (Elena K., 2009). McCarter and Vazquez (1966) explain the distribution of IgM was similar in asthmatic and control lungs . **Results of adenosine deaminase enzyme specific activity are important to give us clear a picture of the human immune defense system . The data of ADA specific activity in sera of the healthy control and the patients group are presented in Table 3 , the specific activity of adenosine deaminase was increased highly significant ($P \leq 0.01$) compared with control group.**

Table 3 : Specific activity of adenosine deaminase in asthmatic patient compared with control

	Adenosin deaminase specific activity
Control	1.4 ± 0.33
Asthma patient	2.9 ± 0.41

ADA enzyme is one of the most essential immune enzyme . Its function gives picture of immune status of the body . It was found to play a critical role in the normal development of the immune system . ADA also essential for proper development of T- and B- lymphocytes in mammals (Hirschorn.,1993) . Adenosine is a signaling nucleoside that accumulates as a result of tissue hypoxia and damage (Hasko and Cronstien .,2004) . Increased levels of this molecule have been found in the lungs of asthmatics (Dreweir *et al.* .,1983) indicating a role in disease pathogenesis. In asthmatic airways, adenosine is a potent bronchoconstrictor with either pro- or anti-inflammatory effects depending on receptors interactions. Adenosine receptors are an essential part of the physiological negative feedback mechanism for limitation and termination of both tissue-specific and systemic inflammatory responses (Fozard ., 2003). In recent clinical studies show, increases in plasma adenosine have been shown to accompany exercise-induced asthma, and adenosine concentrations in exhaled breath condensate are increased in asthmatics. These new data provide support for a key role for adenosine in asthma, which has become increasingly persuasive in recent years .Furthermore, high ADA activity might play a role in the detoxification process of high amount of adenosine substrates , suggesting that adenosine deaminase might play a crucial role as potential novel therapeutics for this condition (Kim *et al.*, 2009).

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تقييم بعض الجوانب المناعية لمرضى الربو
د. ميادة فرحان درويش / كلية العلوم - جامعة الكوفة

الخلاصة :- تناولت الدراسة الحالية تقييم بعض الجوانب المناعية لدى المرضى المصابين بالربو مثل قياس معايير الدم وتشمل العدد الكلي والتفريقي لخلايا الدم البيض وقياس مستوى الـ IgG المناعية (Ig) وقياس فعالية انزيم الـ ADA (ADA) بأعتبره انزيم مناعي ودراسة امكانية استخدام كعلاج مناعي بدلا من العقاقير المستخدمة في علاج مرضى الربو لتجنب التأثيرات الجانبية لبعض الادوية المستخدمة في العلاج ، وتضمنت اختياراً عشوائياً لـ ٣٦ مريض من مستشفى الصدر التعليمي في محافظة النجف بالاضافة الى ٢٤ شخصا "سويا" بوصفهم مجموعة سيطرة . اظهرت نتائج الدراسة حدوث زيادة في العدد الكلي لخلايا الدم البيض لدى مجموعة المرضى وكانت الزيادة عالية المعنوية عند المقارنة مع مجموعة السيطرة ، كما بينت النتائج حدوث زيادة عالية المعنوية في عدد neutrophil and basophil cells وبنفس الوقت انخفاض في عدد Lymphocytes and monocytes cells. دللت النتائج انخفاض في مستوى مقابل زيادة في مستوى الـ E ، وبالوقت نفسه بينت النتائج حدوث زيادة في الفعالية النوعية لانزيم ADA لدى مجموعة المرضى بالمقارنة مع مجموعة السيطرة مما يؤكد دوره في تحويل مادة الـ ADA السامة والتي لها دور اساس في تطور مرض الربو الى ساينوسين وهو مركب غير سام .