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Measurement of Some Hematological and Immunological Parameters in the Sera of Asthmatic Patients

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Abstract

The present study was carried out on (80) adults asthmatic patients (43 male and 37 female), their age from (25 -65) years. Some of them acute episodes while other between episodes interval. They were selected from out patients clinics of AL-Zahra'a hospital for allergy and asthma in Baghdad. All patients were diagnosed according to standard medical criteria as being asthmatic. The results of investigations were compared with those for a total of (20) healthy individual [(12) male & (8) female] with age (25 – 45) years, as a control group, throughout the period extended from (July 2014 till December 2014).

The patient samples were applied to determine complete blood count include (Hb, PCV, RBC count, WBC & Eosinophil count) in addition to (IL-8, GM-CSF) which estimated by ELISA technique. The results revealed that the mean Hb level for asthmatic patients were lower than normal value (9.3 ± 1.4 g/L, 8.7 ± 1.5 g/L) male & female respectively. More ever PCV, RBC, count values for both were nearly equal with in low values (26 ± 4 , 3.6 ± 0.3 male, 25 ± 3 , 3.7 ± 0.35 female).

In this study the patients suffered from Iron deficiency anemia and nutritional deficiency. While leukocyte and eosinophile count values where higher than normal male: (12.5 ± 0.5 , 0.5 ± 0.17), female: (12.6 ± 0.6 , 0.67 ± 0.32) respectively and this due to asthmatic state. Most of sera patients had high concentration of Cytokine (IL-8 & GM-CSF). The values concentration were found to be as high as (177pg/ml , 43 pg/ml) respectively in compared with healthy control (70 pg/ml , 8.2 pg/ml).

Keywords: asthmatic patients, hematological parameters, cytokines.

قياس بعض المعايير الدموية والمناعية في دم ومصل المصابين بالربو

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الخلاصة:

اجريت الدراسة حول (80) مريض (51) ذكر و (29) انثى مصاب بالربو القصبي و بعمر يتراوح ما بين (25-65) سنة منهم في حالة نوبة رئوية حادة وبعضهم في الفترة ما بين النوبات ممن يتوافدون على مستشفى الزهراء للحساسية والربو في بغداد. تم تشخيص المرضى حسب المواصفات الامراضية للربو القصبي من قبل المختصين وتمت مقارنة نتائج الفحوصات التي اجريت على (20) شخص كسيطرة موجبة تتراوح اعمارهم بين (25-45) سنة منهم (12) ذكر (8) اناث خلال الفترة ما بين (تموز 2014 - كانون الاول 2014).

تم فحص المرضى بعناية فائقة ، سريري و اشعة ثم اخذ عينات من الدم لتعيين مستوى (Compleat Blood Count) والذي يتضمن (Hb , PCV , RBC , Eosinophil & WBC) اضافة لمستوى العاملين (IL-8, IL-6,) بواسطة Elisa. وكانت النتائج تشير الى ان مستوى الهيموغلوبين لدى المرضى المصابين بالربو اقل من النسبة الطبيعية ومقارنة بمجاميع السيطرة الموجبة وعلى التوالي الاثناث (8.7 ± 1.5 g/dl)

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والذكور (9.3+1.4g/dl) وكذلك (RBC count , P.C.V.) كانت القيم متقاربة لكلا الجنسين الاثناث (25+3,3.7+0.35) والذكور (26+4,3.6+0.30) . لوحظ ان المرضى يعانون من فقر الدم بسبب عوز الحديد والغذاء اضافة الى الربو . اما بالنسبة للكريات البيضاء والحمضية (WBC & Eosinophile count) لوحظ انها اكثر من المعدل الطبيعي مقارنة بالسيطرة الموجبة وكانت النتائج كالاتي الاثناث (12.6+0.6,0.67+0.32) والذكور (12.5+0.5,0.5+0.17) وهذا يعود لسبب حالة الربو عند المرضى . اما بالنسبة للفحوصات المناعية فكانت النتائج (IL-8 mean value) (177 pg/ml) لمرضى الربو مقارنة بالاصحاء (70pg/ml) بينما كانت نتائج (GM-CSF mean value) كالآتي (43pg/ml) لمرضى الربو مقارنة بالاصحاء (8.2 pg/ml)

Introduction

Asthma is a disease in which the airway become blocked or narrowed .These effects are usually temporary but they cause shortness of breath , breathing trouble and other symptoms [1]. The risk factor associated with the development of asthma has been the focus of many investigations in the last two decades [2]. Although viral respiratory infections are important triggers of acute bacterial exacerbation are frequent problems in the management of asthma [3]. There is number of pathological abnormalities of bronchial system in asthmatic patients.

Asthma prevalence has increased dramatically in many countries over recent decades, demonstrating that environmental exposures play a dominant role in the etiology of this disease [4]. Worldwide, 130 million people have asthma. The prevalence is [8 – 10] times higher in developed countries than in the developing countries and the prevalence is higher in low income groups [5]. In Egypt, the WHO estimated that chronic respiratory disease account for 6.9% of disease and in terms of specific condition, osteoarthritis, injuries and asthmatic bronchitis are the leading causes of disability [6]. There is increasing evidence from observational studies that there are a strong relationship between diet and respiratory disease .for sodium intake an unfavorable association is observed in asthmatics but the intake of fruit and vegetables and fish in more likely to be beneficial [7].

Materials and methods

A. Patients & Control Groups: Eighty adults' asthmatic patients were included in this study. Their age ranged from 25-65 years old they were chosen from the out patients' clinic of Al-Zahra 'a Hospital for Allergy & Asthma. The results of this study were compared with those for 20 adults' apparently healthy volunteers.

B. Methods

-Diagnosis of Asthmatic Patients: Diagnosis of asthma cases was carried out by consultant physician through chest examination & lung function test beside the chest X-ray.

-Laboratory investigations: Blood samples were collected by vein puncture divided into two portions, the first part were transferred into sterile plain tube, centrifuge & separated serum to be used for assessment of (IL-8, GM-CSF) using ELISA Technique.

The remaining of blood were transfused into (E.D.T.A.) tube for haematological parameters, for determination of Hb (by Cyanamethhaemoglobin method), PCV (Micro haematocrite method) RBCs .WBCs .eosinophil count by proper way for counting [8].

In order to find out of the changes in the hematological parameters for asthmatic patients in comparison with control group and estimate the level of cytokines such as (IL-8&GM-CSF) in sera patients in comparison with apparently healthy control.

Results and Discussion:

1. Distribution of the patients according to age group:

The range of patients age was from 16-65 years old the higher occurrence of infectious asthma was at age group 16-25 years [24 (30%)] patients as show in Table-1. This result showed increased disorder of asthma at age range [16-25] year old due to bad behavior for young patients as smoking.

Table 1- Distribution of asthmatic patients' in relation to age

Age	No	%
16-25	24	30%
26-35	16	20%
36-45	18	22.5%
46-55	12	15%
56-65	10	12.5%
Total	80	100%

These results showed increase disorder of asthma of age rang [16-25] years due to the majority of young patients were diagnosed as having asthma because of chest and food allergy particularly to eggs and fish [9]

2-Distribution of patients according to gender:

The result shows that the occurrence of asthmatic patients at age group [16-25] and [26-36] years were higher in male than female due to bad behaviour for male patients as smoking .Although several studies [10] proved that smoking is major risk factor for lung disease and asthma, either cigarettes andnarghile.

Table 2-Effect of gender & age on development of infectious asthma

Age	Males	Females	Total	
	No	No	No	%
16-25	20	4	24	30
26-35	14	2	16	20
36-45	6	12	18	22.5
46-55	6	6	12	15
56-65	5	5	10	12.5
Total	51	29	80	100

At age groups [36-45] years females developed asthma more frequent than males due to suffere from overweight or obesity that obligate the patients to diminish their physical activity & therefore decrease energy expenditure. Some studies proved that obesity is one of the risk factor for asthma [11]. While in other age groups [(46-55) and (56-65) years] the occurrence nearly equal in both sex.

1. Hematological parameters of the studied groups:

Table 3: shows that, although mean Hb values for males were higher than females (9.3 ± 1.14 , 8.7 ± 1.2 g/dl respectively) in comparison with control Group (12.3 ± 1.5 g/dl) these results revealed that both males and females might have signs of anemia But PCV and RBC count values for both males and females were nearly equals, On the other hand, leucocytes and eosinophils, count values were higher than normal values in ccomparison with healthy control [8].

Table 3 – Hematological parameters of asthmatic subjects.

Parameters	Patients group		Control group
	Males No.=51	Females No. = 29	Healthy individual No.= 20
Hb(g/dl)	9.3 ± 1.4	8.7 ± 1.5	12.3 ± 3.6
RBCs ($10^{12}/L$)	3.6 ± 0.30	3.7 ± 0.35	4.5 ± 0.5
PCV (%)	26 ± 4	25 ± 3	38 ± 2
Leucocytes($10^3/L$)	12.5 ± 0.5	12.6 ± 0.6	7.4 ± 0.6
Eosinophil($10^9/L$)	0.5 ± 0.17	0.67 ± 0.32	0.4 ± 0.16

The results of this study demonstrated that concentrations of hemoglobin, PCV and RBCs of patients with asthma were lower than healthy control and this may predispose patients to anemia. On the other hand, the values of white blood cells and eosinophils were higher than control group. As observed in different studies, the known risk factors for the development of pulmonary disease include degree of anemia, lower hemoglobin and higher steady-state white blood cell count [12].

However [13] decrease in hemoglobin concentration with elevation in white blood cells are common among asthma patients in compared with none asthma [13, 14] finally, although, eosinophil cell protect us against parasites and other infectious agents, its high level is also responsible for the allergic reaction and worsen asthma state [14]. However elevated eosinophils count is considered one of diagnosis factors for occurrence of asthma

3. Level of IL-8 & GM-CSF in the sera of the studied groups:

These assays were carried out on the patients sera to estimate the concentrations of cytokines (IL-8 and GM-CSF) in compared with healthy controls. Interleukin- 8 mean values concentrations were found to be as high as (177 pg/ml) in asthmatic patients followed by (70pg/ ml) in healthy control. The last cytokine in this study GM-CSF which was found to be elevated in asthmatic patients groups (mean 43Pg./ ml), while in healthy controls was [mean. 8.2 Pg./ ml).

Table 4- The difference in mean serum interleukin 8 and GM.CSF level (Pg/ml) between the studied groups.

Interleukin's Concentration Pg./ml	Healthy controls Pg/ml	Asthmatics disease Pg. /ml	P Value
Serum IL-8			
Range	7.0 – 170	52 – 326	<0. 001
Mean	70	177	
SD	61	82	
No	20	80	
Serum GM-CSF			
Range	6.1 – 13.8	7.2 – 152	< 0. 001
Mean	8.2	43	
SD	1.8	4.8	
No	20	80	

(IL-8& GM- CSF)are a pro-inflammatory cytokine that play a pivotal role in asthmatic patients , this is true since arising level of these cytokines in the sera of asthmatic patients as shown in Table-4 in comparison with apparent healthy control group. This findings agreed with the finding of increased allergen-provoked release of GM-CSF and IL-8, and the ability to attenuate enhanced Eosinophil with blocking antibodies for GM-CSF, and to a lesser extent IL-8, suggests that these two cytokines are the main contributors to increased eosinophil survival resulting from allergenic stimulation [15]. These results demonstrate that there are increased numbers of activated eosinophils and GM-CSF is increased in patients [16].

Also that's agree with finding: Most of the patients with bronchial asthma or atopic dermatitis show evidence for up-regulated IL-8 protein expression in eosinophils but not in neutrophils, suggesting that an eosinophil-specific cytokine may act in these patients.

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