

Article

Study of the relationship between genetic polymorphism of the IL-17 gene and vitamin D in asthmatic patients in Thi Qar Governorate

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ABSTRACT

This study was conducted at the Biotechnology Research Laboratory, Mazaya University College, from December to June 2022. This study included 150 samples (100 asthmatic patients and 50 healthy controls). The results showed that the highest incidence of asthma was in patients whose duration of illness ranged between (less than 14) years and reached (69%). The lowest incidence was in patients whose duration of illness ranged from (≥ 30) and more, reaching (6%), with a very high significant difference, which reached (0.00). The results of the current study showed the frequency of genotypes and alleles of the IL-17 gene for the two groups of patients and the comparison, as the percentage of genotype frequency (GG) in the comparison group was (46%) and in the group of patients (44%), without a significant difference, where the value of (OR = 1.00) The percentage of genotype frequency (AA) was in the comparison group (16%) and in the group of patients (32%), with a significant difference of (OR=2.09). As for the frequency of the genotype (GA), it was in the comparison group (38%) and the group of patients (24%) and without significant difference, where the value of (OR = 0.66). The current study showed vitamin D levels in both males and females ($24.63 \pm 15.54 / 23.01 \pm 17.21$), respectively, and without significant differences at the level of probability (0.05) (P.Value=0.794).

Keyword: IL-17, asthmatic, Thi-Qar population.

INTRODUCTION

Asthma is the most common chronic respiratory disease, affecting about 30% of children and 10% of adults. In the Western world, it is a global health problem affecting more than 300 million people of all ages and races. It is estimated that 250,000 thousand people die prematurely every year. General Asthma Worldwide ¹ Asthma is a chronic inflammatory disorder of the lungs associated with airway hyperresponsiveness that leads to recurrent episodes of wheezing, shortness of breath, chest tightness and coughing. Asthma is defined by its clinical, physiological, and pathological characteristics. Wheezing is the most common finding from a public health point of view. ²

In recent years, there has been an increasing relationship between asthma and vitamin D deficiency in the blood worldwide. ³ and both ⁴ indicated a relationship between vitamin D deficiency and increased inflammation in asthma and

exacerbation of asthma. And poor control over it. Asthma is a heterogeneous chronic respiratory disease with genetic and environmental components. It is the most common disease in Ireland, affecting about 20% of children ⁵ Vitamin D deficiency is common all over the world, especially among children ⁶ and several medical conditions are associated with low serum levels of 25-hydroxyvitamin D (25OHD), including asthma ⁷. Vitamin D deficiency occurs in asthmatic patients due to exposure to common risk factors such as manufacturing, poor diet, obesity, dark skin pigmentation, and the winter season. And high latitude ⁸. Based on this evidence, Litonjua and Weiss hypothesized that vitamin D deficiency is responsible for the asthma epidemic, and a growing body of literature supports the notion that this vitamin is the cause of asthma, sinusitis, and allergic rhinitis, as vitamin D, regulates a wide range of cells. Immunogenic and non-immune regulate the innate and adaptive immune response and cell growth^{9,10}. Asthma is a multifactorial disease. In addition to environmental exposure, genetic factors significantly impact the onset, severity and treatment of asthma. Polymorphisms in different genes have been shown to influence asthma severity, response, and treatment ¹¹. IL-17F-IL-17A has been found in the airways of asthmatic patients, and its expression level correlates with disease severity. Airway activity and mucus hypersecretion and significantly higher serum levels of IL-17 were found in patients with uncontrolled asthma compared to well-controlled and healthy patients ¹²

MATERIALS AND METHODS

Samples Collection

One hundred blood samples were collected from the Chest and Respiratory Center in Thi Qar Governorate and some private medical clinics in Thi Qar Governorate for people with asthma, representing the experimental group. Furthermore, 50 blood samples for healthy people (students and self-employed people of both sexes), where an amount of (5 ml) of venous blood was taken from the two groups of healthy and sick people, and (2 ml) of blood samples were placed in tubes containing EDTA, an anticoagulant, and stored at -20°C. Also, (2) ml of blood was placed in Gel tubes to measure and compare the amounts of IL-17 and Vitamin D in healthy and sick patients. An information form was approved for the two groups of patients and healthy people, including (vitamin D percentage, age, sex, smoking, area of residence, duration of infection, and family history).

DNA Extraction

DNA extraction from the patient and healthy samples included several steps based on the leaflet attached to the kit DNA Extraction manufactured by Geneaid (Korean origin).

Polymerase Chain Reaction (PCR)

PCR technique was used to amplify the (IL-17) genes according to the method of work recorded (Arand et al., 1996).

Primers

PCR was used to amplify the (IL-17)gene using specific primers. Primer

F 5- ACCTGGCCAAGGAATCTGTG-3

R 5- GCAAGAGCATCGCACGTTAG-3

which is designed in this study to amplify 526 bp.

No.	Steps	Temperature	Time	No. of cycle	Size (bp)
1	Initial denaturation	95	10 m.	1	526
2	Denaturation	95	35 sec.	30	
	Annealing	62	35 sec.		
	Extention	72	35 sec.		
3	Final extension	72	10 m.	1	

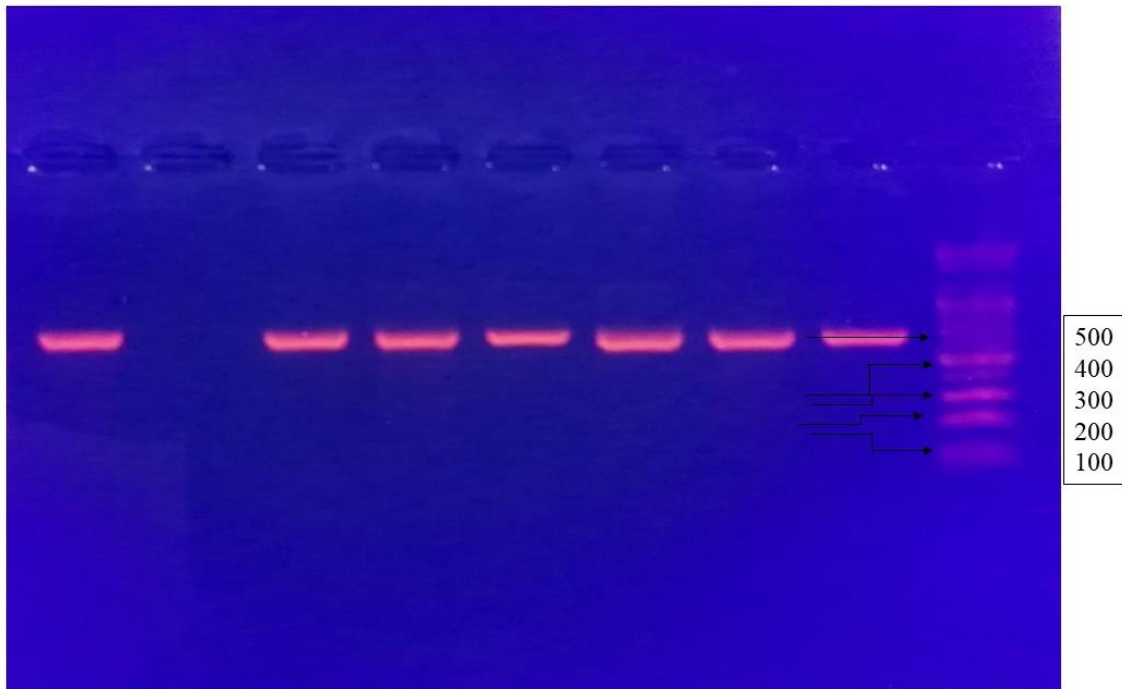


Figure 1. Show Electrophoresis of PCR products on 2% agarose gel. IL-17 gene and bundle appearance at the base pair 526 bp

RESULTS

Location	Patient Group %	Control %
Rural	21	16
Urban	79	34

P- value = 0.14

Table 2. Distribution of the two groups of patients by area of residence for both sexes

Location	Patient Group %	Control %
Male	30	28
Female	70	72

P- value = 0.80

Table3. Distribution of control groups and patients by sex

Location	Patient Group %	Control %
Smoking	14	5
Non-Smoking	86	45

P- value = 0.78

Table 4. Distribution of the two groups of patients and control, according to smoking

Location	Patient Group %	Control %
Vit. D	22.50 ± 13.35	17.72±9.78

P-value = 0.301

Table 5. Distribution of control and patient groups according to Vit.D

Genotype	Vit.D
GG	22.20± 14.22
GA	21.30± 11.10
AA	21.18± 12.09

P-value = 0.97

Table 6. Vitamin D level according to the genotypes of the IL-17 gene (-197 G>A) in the two groups of patients and the comparison

95% CI	OR	Patient Group N=100 (%)	Group Comparison (%)N=50	Genotypes
—	1.00	(% 44) 44	(% 46) 23	GG

1.44 - 0.30	0.66	(% 24) 24	(% 38) 19	GA
5.27 - 0.82	2.09	(% 32) 32	(% 16) 8	AA
		(%100) 100	(%100) 50	the total
Allele frequency				
————	1.0	(% 56) 112	(% 65) 65	G
2.39 - 0.88	1.45	(% 44) 88	(% 35) 35	A
		(%100) 200	(%100) 100	the total
OR: Odd Ratios	95% CI Confidence Interval			

Table 7. Frequency of IL-17 genotypes for both patient and comparison groups

DISCUSSION

The relationship between vitamin D and the IL-17 gene polymorphism in patients with Asthma

Asthma is a chronic disease that affects adults and children. It includes many symptoms resulting from exposure to various factors and causes. It is widespread in all countries of the world. Epidemiological studies have shown that asthma affects 20% of the population of any country in the world and is more prevalent in Western and Asian countries.¹³ The incidence of asthma has increased, and according to the statistics of the World Health Organization, the incidence of asthma increased by 50% every ten years¹⁴. The prevalence of asthma varies between geographical regions, and this may be due to different social habits, lifestyles and other factors¹⁵. The prevalence of asthma in urban areas (79%) et al., (2011) The proportion of patients from urban areas reached (80%) and (20%) are residents of rural areas. As for the study published by¹⁶, it was found that the spread of the disease was transmitted between coastal areas, small areas and small areas, which amounted to 7.7%). In comparison, the areas were (6.7%) The results of the current study may explain the high incidence of infection in urban areas that living in cities is a risk factor for the occurrence of asthma, which may be due to the lifestyle and the huge increase in the number of cases. Cars that contribute to air pollution¹⁷ and Weitzman et al., 1990 The results of the current study also showed in Table 3 that there are significant differences concerning sex, as females were more affected than males by (70%), while males (30%) and this is also what was indicated by¹⁸, where they found that the percentage of infection The infection of females (60%) and males (40%), as well as the study conducted by¹⁹, found that the percentage of females with asthma is (62.6%), while the percentage of males was (37.4%)and between (Bush & Menzies-Gow, 2009) The difference in incidence rates during life stages may be due to the hormones that affect the incidence and severity of asthma²⁰. It was found that asthma is more frequent in females after the onset of menstruation²¹. Also, the results of the current study showed in Table 4 that the majority of patients are non-smokers (86%), while the percentage of smokers is (14%). However, smoking is a risk factor for the occurrence of asthma, as the exacerbation of the disease occurs more in smokers because smoking leads to a Severe decline in

lung function²² and Bai et al., 2007 The results of the current study showed that the levels of vitamin D in the group of patients were 22.50 ± 13.35) and in the comparison group (17.72 ± 9.78) and without significant differences at the level of probability (0.05) (P.Value=0.301) and (T-value=1.04), as shown in Table (5).

CONCLUSION

The results of the study showed a relationship between mutations and polymorphisms in the IL-17 gene and asthma, and the polymorphism, deletion and insertion had an important role in the pathophysiology of asthma and its relationship to vitamin D. The study recorded some mutations in the clinical variation site at the National Center for Biotechnology Information of NCBI. The current study also indicated that mutations affecting the encoded genes and low vitamin D could be a direct cause of asthma.

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