

Original Research Article

Frequency of HLA-DR3 Alleles and Pedigree Analysis for Patients with Thyroiditis

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Abstract

A study was performed to estimation the concentration of HLA-DR3 alleles and pedigree analysis in patients with thyroid diseases in Babylon province .The results showed that there was significantly increased ($p<0.05$) in frequency of HLA-DR alleles in all groups of patients (hyperthyroidism , hypothyroidism , euthyroidism and normal with family history) compared with healthy subjects . Furthermore , the results reveals increased ratio of disease in females compared with males , also the ratio of thyroiditis patients were higher in second age categories compared with first and third age categories . The pedigree analysis showed genetic role in the frequency of disease depending on the genetic history of family , where more patients with thyroiditis were found in families, one of them (especially mother) or their uncles undergo from this disease .

Key words: Autoimmunity . Thyroiditis . HLA-DR Alleles.Pedigree analysis

الخلاصة

صممت هذه الدراسة لتقدير تركيز أليلات مستضد خلايا الدم البيض وشجرة تحليل النسب لمرضى التهاب الدرقية في محافظة بابل .وأظهرت النتائج وجود زيادة معنوية في تكرار تلك الأليلات لكل مجاميع المرضى (فرط إفراز الدرقية ، قلة إفراز الدرقية ، متلازمة أسوياء الدرقية و مجموعة التاريخ العائلي) مقارنة مع الأفراد الأصحاء ، إضافة الى ذلك بينت النتائج زيادة نسبة المرض في الإناث مقارنة مع الذكور ، كذلك كانت نسبة مرضى التهاب الدرقية أعلى في الفئة العمرية الثانية مقارنة مع الفئات الأخرى . بينت شجرة تحليل النسب الدور الوراثي في تكرار المرض اعتماداً على التاريخ الوراثي للعائلة ، وان أكثر المرضى قد لوحظوا في العوائل التي أحد أبويهم (خصوصاً الأم) أو أعمامهم يعانون من ذلك المرض .

Introduction

The immune response to self-antigens or antigens associated with the commensal microbiota are called autoimmunity and can lead to autoimmune diseases that are characterized by tissue damage [1,2]. In organ-specific diseases, autoantigens from one or a few organ only are targeted, and disease is therefore limited to those organs for examples of organ specific autoimmune diseases are Hashimotos' thyroiditis and Graves'

disease, both predominantly affecting the thyroid gland [3]. The mechanism of Graves' disease characterized by production of autoantibodies against the thyroid-stimulating hormone receptor(TSHR), and the consequences is hyperthyroidism which means overproduction of thyroid hormones (T3,T4 and TSH) [4,5], moreover this disease is discovered mostly in the third and fourth decades of life[6].The Hashimotos' disease was represented by production of thyroglobulin (Tg), thyroid

peroxidase (TPO) autoantibodies and autoreactive T-cells against thyroid antigens, and the results is destruction of thyroid tissue leading to hypothyroidism (underproduction of thyroid hormones) [7, 8]. Studies have shown the existence of power correlation between HLA gene and its specific autoimmune disease and this is expressed by relative risk, in addition, there is a different in the recurrence of HLA-DR alleles in patients with thyroiditis within population groups according to the ethnicity and geographic location [3, 9]. Therefore, the aim of this study to determine the concentration of HLA-DR3 alleles and pedigree analysis, as well as their relationship with age and sex, in order to knowledge the correlation between thyroiditis with environmental and genetic factors in Babylon province.

Materials and Methods

1- Patients and Control

The work was applied on 60 patients of thyroiditis (12 males and 48 females) admitted to the Teaching Hilla hospital and General Mehaweel Hospital in Babylon province, and 21 apparently subjects (7 males and 14 females) with no symptoms of thyroiditis were selected as control group. Patients were classified into four subgroups: hyperthyroidism (20 patients), hypothyroidism (20 patients), euthyroidism (10 patients) and normal with family history (10 patients), while the cases of this study were divided into three age categories for comparison: (6-25), (26-45) and (46-65) years. The number of cases was 15, 31 and 14 in each category respectively.

2- Blood Samples

The blood samples were drawn from each patients and control (5ml) by vein puncture using disposable syringes. The blood was placed in the Jel tube and kept to clot at room temperature (25-20) ° C, then centrifugated at 3000 rpm for 10 minutes after that sera samples were carefully transferred to Eppendorf tubes and preserved at -20° until use [10].

3- Immunological Test

The frequency of HLA-DR alleles was estimated by ELISA according to the manual procedure of DAI company (USA).

4. Pedigree Analysis

Pedigree was drawn from the information obtained from the sons about their parents, and the parents about their grandfather and grandmother; therefore, according to the information that has been re-establishment the genetic history of the personal qualities [11].

5- Statistical Analysis

The results were analyzed using statistical system SPSS version -17 (T-testing and ANOVA-LSD-General Linear Model).

Results

The mean level of HLA-DR3 alleles showed that a significant differences ($P = <0.05$) in hypothyroidism patients when compared with hyperthyroidism, euthyroidism and control groups, furthermore they revealed significant differences in the level of this allele in family history patients, when compared with hyperthyroidism, euthyroidism and a healthy individuals groups, as illustrated in table (1).

Table 1: The concentrations of HLA-DR3 in patients with thyroiditis

Groups Parameter	Control ±S.E Mean N=21	Hyperthyroidism Mean ± S.E N=20	Hypothyroidism Mean±S.E N=20	Euthyroidism ±S.E Mean N=10	Family history Mean ±S.E N=10	L.S.D
HLA-DR3 IU/ml	12.55±0.79	16.64± 1.35	22.33±2.10	± 2.29 17.46	21.24 ± 2.72	3.8

The result showed that the ratio of patients with thyroiditis were higher in females compared with males in all age categories, which reached 66.67, 87.1

and 78.57 % in the first, second and third age groups respectively, while the ratio in males were 33.33, 12.9 and 21.43 % consecutively(Figure-1).

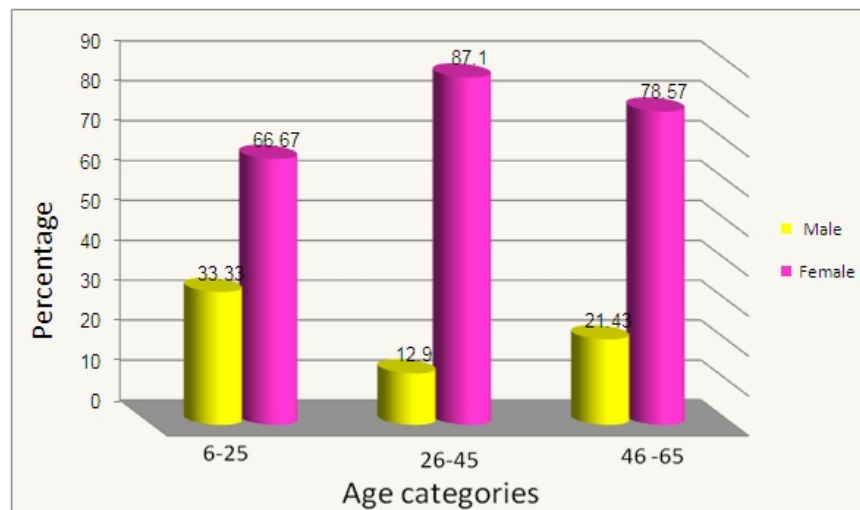


Figure 1: The relationship between sex and patients with thyroiditis of different age categories .

The study showed that there is a genetic predisposition in the frequency of thyroiditis in patients that their families undergo from this disease .The data showed that the ratio of patients who inherit thyroiditis were higher when their mothers undergo from this disease (30%), while 20% of the cases studied

inherited from their father. Furthermore , the percentage reached 23.33% and 15% when the disease were frequented in their uncles from mother and father respectively , but this percentage was 11.67% in patients that not inherited the disease , as illustrated in figures 2 and 3 (A, B, C).

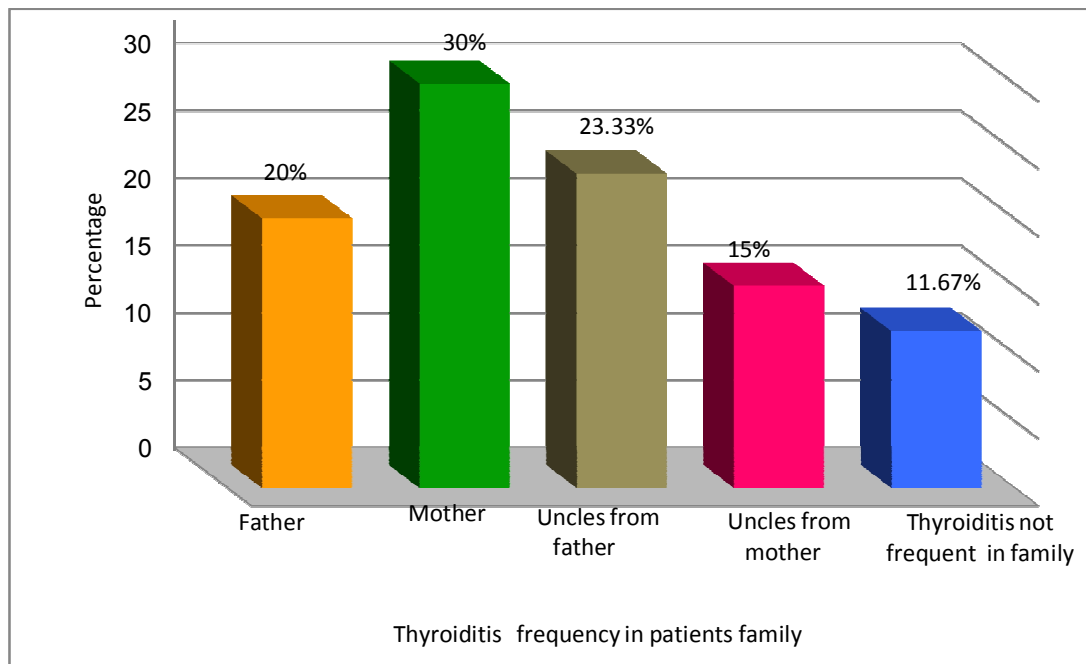


Figure 2: Frequency of thyroiditis in families of patients.

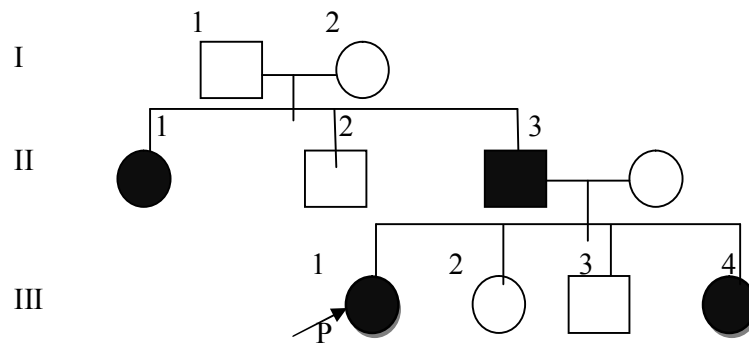
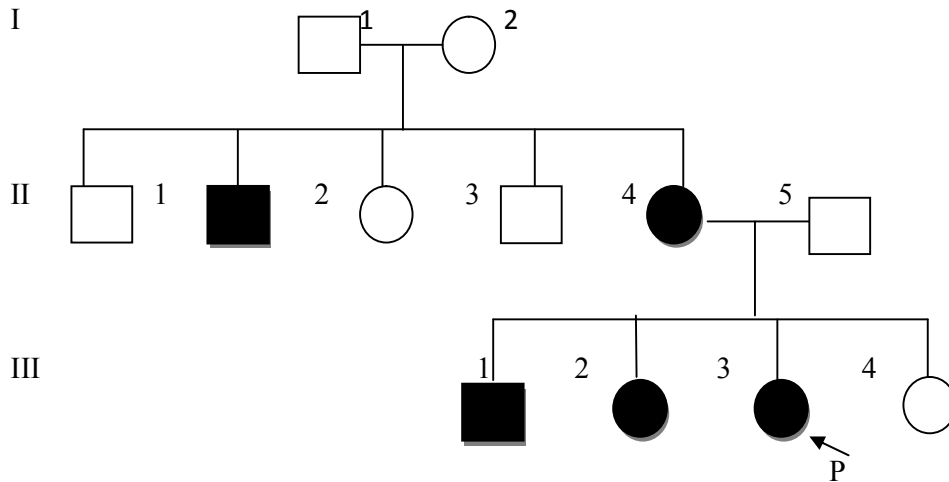
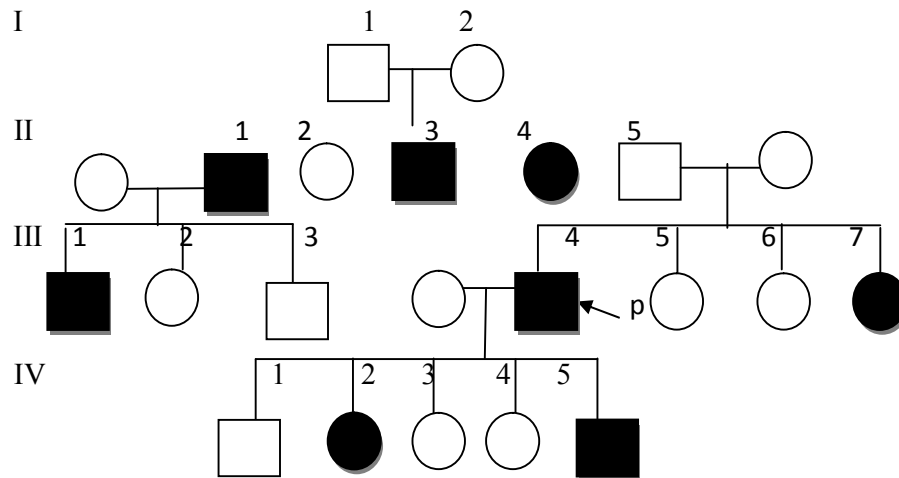


Figure 3 A : pedigree analysis showing the frequency of thyroiditis in family members (III-1) for three generations their father was infected .



B : pedigree analysis showing the frequency of thyroiditis in family members (III-3) for three generations their mother was infected .



C : pedigree analysis showing the frequency of thyroiditis in family members (III-4) for four generations their uncles was infected .

Discussion

The patients showed that a significant differences in the concentration of HLA-DR3 in all groups of patients in comparison with healthy subjects. [9] revealed that the HLA region is a highly polymorphic region that contains many immune response genes and has been found to be associated with various

autoimmune disorders. In Hashimoto's thyroiditis, aberrant expression of HLA class II molecules on thyrocytes has been demonstrated, presumably; such thyrocytes may act as APCs capable of presenting the thyroid auto antigens and initiating autoimmune thyroid disease[12,13], while in graves' disease the HLA-DR3 was reported by several

studies as primary susceptibility allele, no consistent associations were observed in Hashimoto's thyroiditis. In Caucasians, associations of different forms of HT with various HLA alleles were reported, including DR3 [14], DR5, DQ7 [15], DQ B1*03 [16], DQw7 [17] or DRB1*04-DQB1*0301 haplotype [18].

The results showed that the ratio of incidence of disease increased in females compared with males in all age groups. [19] and [20] reported that a high frequency of disease in women when compared with men [21, 22, 23, 24]. Thyroid disorders are prevalent worldwide, especially in women, and this is associated with sex hormones and the X chromosome which affect the thyroid and immune system [25].

Results revealed recurrence of thyroid disease in the families of patients. [26] reported that the molecular characterization of a case of severe congenital hyperthyroidism with a history of hyperthyroidism in the paternal aunt and the paternal grandmother, who were both found to be heterozygous for a mutation located in exon 10 of the TSHR gene. [27] studied genetic analysis and family pedigree for boy suffering from hypothyroidism and carries an unbalanced translocation t(8;16), and was found in other three phenotypically normal family members (the patient's mother, one maternal uncle, and the maternal grandfather). Furthermore, [28] has shown the segregation analysis co-dominant Mendelian inheritance provided a significantly better fit to the data than either dominant or recessive inheritance. In other words, genetic components involved in autoimmune thyroid diseases (AITDs) have additive effects until they reach, together with environmental and endocrine factors, a threshold allowing occurrence of the disease. Meantime, [29] reported that more than 30% of the patients who suffer from the Graves' disease and Hashimoto's thyroiditis have a family history for disease. In fact, many candidate genes involved in both immune process

and thyroid physiology have been investigated and have shown different degrees of involvement in AITDs pathogenesis [30-32].

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