

The Association of Myasthenia Gravis with HLA class II Antigenes in Iraqi Patients

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Abstract:

The nature and intensity of the association of myasthenia gravis (MG) with distinct human leukocyte antigen (HLA) haplotypes differ between ethnic populations, so this study determined the association of HLA class II antigens with myasthenia gravis (MG) in Iraq. The study included Iraqi patients diagnosed with MG and two control groups the first of 54 insulin dependent diabetes mellitus patients and the second of 237 subjects as a normal control group. The test used was microlymphocytotoxicity test. The work was done in the Teaching Laboratories/Medical City/Baghdad. Results: positive associations were observed (etiological risk factors) as follows:

1. HLA-DR locus showed one positively associated allele when compared to healthy control and this was *HLA-DR3* (RR: 21.05, EF 0.73, & P value < 0.05), While when compared to IDDM control no significant association appeared (since the same allele is positively associated with IDDM).
2. HLA-DQ locus showed only one positively associated allele when compared to healthy control; this was *HLA-DQ2* (RR 4.67, EF 0.50, and P value < 0.05). While no significant association appeared when compared to IDDM control.

Other important clinical associations were observed; association with age, gender, strong stressful events, thymoma, and other autoimmune disorders.

Conclusion: The positively associated antigens which were found as follows HLA-DR3 and HLA-DQ2, while no negative association was detected.

Keywords: myasthenia gravis, HLA antigens, major histocompatibility complex.

Introduction:

Myasthenia gravis (MG) is a disorder of the neuromuscular junction resulting in a pure motor syndrome characterized by weakness and fatigue particularly of the extraocular, pharyngeal, facial, cervical, proximal limb and respiratory musculature. Typical and neonatal forms are immunologically mediated. A number of congenital forms of obscure pathogenesis exist. Onset may be sudden and severe (myasthenic crisis) but, more typically, is mild and intermittent over many years. [1, 2] There is a predilection for the external

ocular muscles and certain other cranial muscles, including the masticatory, facial, pharyngeal, and laryngeal muscles. Respiratory and limb muscles may also be affected. [3,4] The disease could affect persons at any age group, but the usually affected patients lie between the ages of 15-50 years old and women affected more than men. [5] There are different autoimmune disorders associated with myasthenia gravis. Different HLA class II antigens (the expressed products of the major histocompatibility complex genes;

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MHC class II) have been associated to the disease as an etiological risk factor.[6,7,8]

Patients and methods:

The study involved 30 Iraqi MG patients of ages 10-72 years old (50% of them were females). Patients included in this study were diagnosed as having definite MG by neurology specialists at three teaching general hospitals in Baghdad (Baghdad, Kadhimya, and Yarmouk teaching hospitals). The sample collection period extended from March 2006 to may 2007. Apparently healthy control of 237 subjects (aging from 4-63 years old, 120 males and 117 females), and 54 insulin dependent diabetes mellitus patients as autoimmune patient control (age ranging 3-75 years old, 24 males and 30 females) were also included in the study. The procedure done to analyse HLA typing by microlymphocytotoxicity test. [3]

Results:

General aspects and clinical manifestations:

By assessing the results obtained from patient and control groups; the following results observed: The mean age for males was 43.5 and females was 29.9 years old; 30% of patients 21-30 years of age, 26.6% 31-40, 16.6% under 20 years old, 16.6% 41-50, 6.6 over 61 years old, while only 3.3 between 51-60 years of age as , and male to female ratio was 1:1; shown in (table-1)

Table -1 Distribution of cases by age and gender.

Age of the patient	Male no. (%)	Female no. (%)	Total no. (%)
< 20	1 (6.6)	4 (26.6)	5 (16.6)
21 – 30	5 (33.3)	4 (26.6)	9 (30)
31 – 40	5 (33.3)	3 (20)	8 (26.6)
41 – 50	2 (13.3)	3 (20)	5 (16.6)
51 – 60	0 (0)	1 (6.6)	1 (3.3)
> 61	2 (13.3)	0 (0)	2 (6.6)
Total	15 (100)	15 (100)	30 (100)

The association with other autoimmune disorders:

In 20% of patients (5 cases) other autoimmune disorders diagnosed by specialist doctors at the medical city were associated with MG and as shown in table-2.

Table -2 Autoimmune diseases associated with MG in this study.

The autoimmune disease	Number of cases (%)	
Insulin dependent diabetes mellitus	2	40%
Rheumatoid arthritis	1	20%
Uveitis (Behcet disease)	1	20%
Addison's disease	1	20%
Total	5	100%

Thymoma was found to be associated with the diseases in 5 patients (17% of the cases).

Strong stressful event was found to precede the onset of the disease in 30% of cases (9 patients) as shown in figure-1.

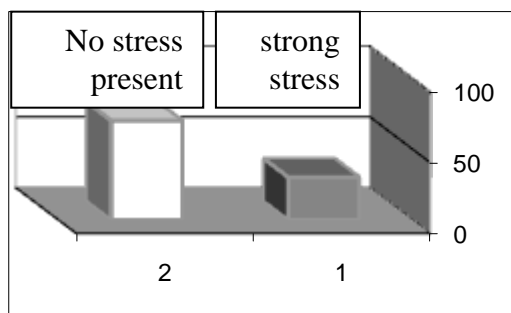


Fig-1 Association of stress with onset of the disease.

Table-3 Distribution of patients by presenting symptom at onset of the disease.

Presenting Symptoms	Male no. (%)	Female no. (%)	Total no. (%)
Diplopia	7 (46.6)	2 (13.3)	9 (30)
Ptosis	4 (26.6)	5 (33.3)	9 (30)
Dysphagia	4 (26.6)	1 (6.6)	5 (16.6)
G. weakness	0 (0)	4 (26.6)	4 (13.3)
UL weakness	0 (0)	2 (13.3)	2 (6.6)
LL weakness	0 (0)	1 (6.6)	1 (3.3)
Total	5 (100)	15 (100)	30 (100)

For HLA class II antigens, the following results were observed (table-4):

1. *HLA-DR locus* showed one positively associated allele when compared to healthy control and this was *HLA-DR3* (RR: 21.05, EF:0.73, PC: 7×10^{-8}), while when compared to IDDM control no significant association appeared (since the same

allele is positively associated with IDDM).

2. *HLA-DQ locus* showed only one positively associated allele when compared to healthy control; this was *HLA-DQ2* (RR: 4.67, EF: 0.50, PC: 2×10^{-4}) While no significant association appeared when compared to IDDM control.

Table 4. Antigens of HLA system showing significant variation between MG patients and Healthy controls.

HLA Antigens	Myasthenia Gravis (No. = 30)		Healthy Controls (No. = 237)		*RR	*EF	*PF	*P	*PC
	No.	%	No.	%					
DR3	23	(76.67)	32	(13.50)	21.05	0.73	-	1×10^{-8}	7×10^{-8}
DQ2	19	(63.3)	64	(27.0)	4.67	0.50	-	1×10^{-4}	2×10^{-4}

*RR= relative risk, EF= etiological fraction, PF= preventive fraction, P= probability, PC= corrected P.

Discussions:

The minimum age of onset observed in this study was 8 years old, while the maximum age of onset was 66 years old with the highest onset at age group 21-30 years old, while the mean age for males was 43.5 years old and for females was 29.9 years old, which is slightly similar to the results recorded by other studies which mentioned that the mean age of onset of the disease for females is between 10-30 years and for males between 60-80 years, [4,7,9] The overall ratio of female: male in this study was 1:1, while most studies mention that this female: male ratio is 3:2, [5,8,10] This difference may be due to the limited number of cases (30 cases) involved in this study and the difficulties in attending Baghdad specialized hospitals from different governorates especially for female patients due to the present improper security situation and difficulty in travelling, also many patients prefers attending private clinics. The presenting symptoms at onset were as follows; diplopia and ptosis were the highest presenting symptoms with percentage of 30 % for each, followed by dysphagia (16.6 %) then generalised weakness (13.3 %), upper extremity weakness (6.6 %), and lower extremity weakness (3.3 %), respectively, compared to other studies it has been mentioned that diplopia is the first in (41%) followed by ptosis (25 %), dysarthria (16 %), lower extremity weakness (13 %), generalised weakness (11 %), dysphagia (10 %), upper extremity weakness (7 %), and masticatory weakness (7 %). [1, 7, 8, 11] The presence of thymoma were observed in 17 % of cases, which is close to that observed by other studies which mention 10-15 % of patients had true thymic tumour. The presences of other autoimmune disorders were confirmed in 20 % of

cases, compared to about 10 % association mentioned by other studies. [7, 12, 13] Strong stressful events were found to precede the onset of the disease in 30 % of cases, and this might point to the effect of psychological stress on the immunoregulatory mechanisms.

Association of myasthenia gravis with HLA typing (class II antigens):

An important factor which plays a significant role in the susceptibility to myasthenia gravis is HLA genotype. [3]

When comparing the results of this study to that of other countries and different ethnic groups the following observed:

1. HLA-DR locus: many associations exists, *HLA-DR2* and *HLA-DR3* were recorded in North Europeans, *HLA-DR3* in Turkey, and *HLA-DR4* in elderly Caucasians who were *HLA-DR3* negative. [14,15,16,17,18,19]

2. HLA-DQ locus: In Jamaicans, *HLA-DQ4* was positively associated, *HLA-DQB1* in Japan, in Sweden *HLA-DQ2* was positively associated with MG, *HLA-DQ6* is negatively associated, also *HLA-DQ8* observed in elderly Swedish patients. [20,21,22,23,24]

Conclusion:

The HLA class II typing revealed that, the following positive associations (as an etiological risk factors) were observed in Iraqi patients; *HLA-DR3*, and *HLA-DQ2*.

This study confirms the heterogeneity of MG in HLA typing among different ethnic groups and pointing to different type of sensitive genes detected in our community.

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مرض الوهن العضلي الوبيل ومرافقة مستضدات الخلايا البشرية البيضاء الصنف الثاني لمرضى عراقيين

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الخلاصة:

ان طبيعة وشدة الترافق بين مرض الوهن العضلي الوبيل ومرافقة مستضدات الخلايا البشرية البيضاء من الصنف الثاني تختلف بين المجموعات الاثنية المختلفة، لذلك فان هذا البحث يهدف لمعرفة الترافق بين مستضدات الخلايا البشرية البيضاء ومرض الوهن العضلي الوبيل للمرضى العراقيين. شملت هذه الدراسة ثلاثين مريضا مشخص بالوهن العضلي الوبيل ومجموعتين ضابطة، الاولى مكونة من 54 مريضا مصابا بدا السكري المعتد على الانسولين والثانية 237 كمجموعة ضابطة من الاصحاء. اجري الفحص بطريقة Micrlymphocytotoxicity. تم انجاز العمل في المختبرات التعليمية بمدينة الطب. النتائج: تم ايجاد علاقة مرافقة ايجابية (عامل مسبب للخطورة) وكالاتي:

1. موقع HLA-DR اظهر ترافق ايجابي في الليل واحد عند مقارنته بالمجموعه الضابطة من الاصحاء وهو HLA-DR3 (الخطر النسبي: 21.05 ومعامل مسبب الخطورة 0.73 ومعامل الاحتمالية > 0.05)، بينما عند المقارنة مع المجموعة الضابطة من مرضى داء السكري المعتمد على الانسولين لم تظهر اي مرافقة ذات قيمة معنوية من الناحية الاحصائية (لان نفس الاليل يحمل مرافقة موجبة مع داء السكري المعتمد على الانسولين)
2. موقع HLA-DQ اظهر ترافق ايجابي في الليل واحد عند مقارنته بالمجموعه الضابطة من الاصحاء وهو HLA-DQ2 (الخطر النسبي: 4.67 ومعامل مسبب الخطورة 0.05 ومعامل الاحتمالية > 0.05)، بينما عند المقارنة مع المجموعة الضابطة من مرضى داء السكري المعتمد على الانسولين لم تظهر اي مرافقة ذات قيمة معنوية من الناحية الاحصائية. تم ملاحظة مترافقات سريرية اخرى : كالترفق مع العمر والجنس، والضغط النفسي الشديد، وورم الغدة الزعترية وامراض المناعة للذات الاخرى.