Anti-Myelin Associated Glycoprotein Antibody and Relapsing Remitting Multiple Sclerosis among A Sample of Iraqi Patients

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Abstract

Background: Multiple sclerosis (MS) is an inflammatory demyelinating disease of the central nervous system affecting nearly 2 million people worldwide. The pathogenesis of MS is at best incompletely understood. There are several proposed mechanisms that may be important in the production of MS plaques: autoimmunity, infection, bystander demyelination and heredity. Deviation of immune responses plays a central role in the pathogenesis of MS. Auto antibodies to many myelin constituents is present in MS. These antibodies may be an autoimmune reaction to an "MS antigens" or simply part of a generalized "non-sense" antibody response. In either case, the antibodies may affect the pathology of MS.

Objective: To study the association between Anti-myelin associated glycoprotein (anti – MAG) antibody and relapsing remitting multiple sclerosis in Iraq.

Method: 105 patients with relapsing remitting multiple sclerosis and 85 patients with other neurological diseases (OND) as control group were recruited in this study. The cases were collected from Baghdad teaching hospital, MS center, Baghdad, Iraq at the period from April 2009 to February 2010. The sera of all patents were tested for MAG- antibodies.

Results: The present study showed increase in the percent of patients with positive anti-MAG antibody in patients with relapsing remitting multiple sclerosis than the control group (7.6% in the patients group viz 3.5% in the control group) but with no statistical significance. Also among patients group we try to study the relationship between severity of the disease measured by expanded disability status scale (EDSS) and positive serum anti-MAG antibody and we found no significant statistical association.

Conclusion: There is a higher percent of patients with positive anti-MAG antibody in the serum of patients with relapsing remitting multiple sclerosis than the control group.

Keywords: Expanded Disability Status Scale, relapsing remitting multiple sclerosis, Anti-myelin associated glycoprotein, immunofluorescence assay.

Introduction

Multiple sclerosis (MS): is an inflammatory disease in which the fatty myelin sheaths around the brain and spinal cord are damaged, leading to demyelination and scarring as well as broad spectrum of signs and symptoms [1]. Disease onset usually occurs in young adults, and it is more common in women. It has a prevalence that ranges between 2 and 150 per 100.000 [2]. MS was first described in 1868 by Jean- Martin Charcot [3]. The main pathologic hallmark of MS is the demyelinatig plague, which has specific histological and immunological characteristic depending on the severity of the disease [4, 5, 6, 7]. Another important

immunopathological feature of MS is continuous synthesis of immunoglobulin (oligoclonal bands) in cerebrospinal fluid CSF [8]. For many years, self reactive anti bodies have been associated with the pathogenesis of MS, and their presence based on studies done in experimental auto immune encephalomyelitis, the most commonly studied model in MS [9].

Myelin associated globulin (MAG) has five (immunoglobulin-like) domains exposed extracellular, makes it accessible to antibodies [10]. The presence of elevated serum anti-myelin antibodies may predict progression from a clinically isolated syndrome (CIS) to multiple sclerosis, and may also correlate with disease activity, but the data are conflicting [11]. The role of B cells in MS is complex. It may act as antigen presenting cells [12] or as cytokine producing cells [13]. Antibodies play a deleterious role when bound to myelin and result in its degradation [14]. Alternatively antimyelin antibodies clear myelin depris from sites of acute degradation to promote remyelination. [15].

High concentrations of IgM (MAG) auto antibodies are found in approximately 50% of patients with peripheral neuropathies accompanied by IgM monoclonal gammopathies. Lower concentrations of MAG IgM autoantibodies can also be found in patients with inflammatory neuropathies, multiple sclerosis and healthy individuals [16]. Detection of IgM autoantibody against myelin associated glycoprotein (MAG) suggests active demyelination in peripheral neuropathy [17]. Detection of low levels of Anti-MAG antibodies in MS patients suggest that there is a low level of humoral immunity to MAG in MS patients that can only be detected by the most sensitive assays. This weak immune response to MAG may be secondary to the demyelinating process, but could play a role in the progression of the disease [18]. In this study we try to find an association between Anti- MAG and relapsing remitting multiple sclerosis, such an association could be of great concern for both neurologist and other health givers in such field.

Patients and Methods

This is a case – control, study included 105 patients with relapsing remitting multiple sclerosis (RRMS) 36 males and 69 females, their age ranged between 20 and 52 years with a mean of (36.45± 5.3). They were recruited from the multiple sclerosis clinics at Baghdad teaching hospital, Baghdad, Iraq. Cases were collected during the period from April 2009 to the end of January 2010. Inclusion criteria included cases of relapsing remitting definite MS who were diagnosed according to the famous MacDonald criteria which is applied in above Baghdad MS clinic. A control group of eighty five individuals (36males and 49 females) with other neurological diseases (ONDS) were also enrolled in this study, their age ranged from 20 to 51 years with a mean of (33.21± 8.2). They were recruited from the consulting clinic of neurology and they were presented with other neurological disease like: epilepsy, migraine and tension type headache. A written consent from the patients were taken.

Base line data were obtained from the patients according to the questionnaire forma that is prepared for this purpose. Severity were calculated according to the expanded disability scale status (EDSS).
Immunofluorescence assay kit for detection of IgG auto-antibodies against myelin sheaths and myelin associated glycoprotein (anti-MAG) in serum was used. (The Binding Site Group, Ltd, Birmingham, UK, Catalogue Number MAG 1567 was used).

The test was performed in immunology department of teaching laboratories, Baghdad medical city, following the procedure protocol included within the kit packing as issued from the manufacturer company.

Statistical Package for Social Sciences (SSPS) version 15 was used for data entry and analysis. Results were expressed in simple statistical terms such as means, percentages, and standard deviations. Exact fisher test was used for testing the significance of association between two discrete variables. Finding of P value less than 0.05 was considered significant.

Results

The study included 105 patients with clinically definite multiple sclerosis with relapsing remitting course attended MS clinic in Baghdad teaching hospital in the medical city complex and 85 individuals with other neurological diseases recruited from neurological outpatient clinic of the same hospital served as control .

The patients group comprised 69(65.7%) females and 36(36.3%) males having RRMS. There was a female predominance with female: male ratio = 1.9:1. The control group comprised 49(57.6%) females and 36(42.4%) males, there was female predominance with female: male ratio = 1.3:1

The age of the patients with multiple sclerosis was ranged between 20 and 52 years. The age of patients in the control group was between 21-50 years. The ages of the patients and the control groups were classified into three groups: 20-30, 31-40 and more than 40. T test revealed no significant change between patients and the control groups. P value was > 0.05. As mentioned in Table (1).

The results of immune florescence assay (IFA) test for IgG anti-MAG of study groups was: positive in 8 (7.6%) of the patients group and 3 (3.5%) of the controls group. This study revealed higher percent of positive anti MAG among the patties group than the control group but statistically was not significant as shown in table 2. (P value is >0.05).

The results of seropositive patients (positive ani- MAG antibodies), according to severity of the disease using the EDSS were: (6.2%) in the patients With EDSS ≥ 3.5 and (8.2%) in the patients with EDSS < 3.5 and this result again revealed no significant change between the patients and the control group. P value was > 0.05 as mentioned in Table (3)

The results of seropositive patients according to state of the patient at presentation (with relapse or without) were: (7.1%) in the patients with relapse and (7.8%) in the patients without relapse and again there was no significant difference between the two groups of the study cases (P value is > 0.01), as mentioned in Table (4).

Discussion and conclusion

B cell activation and antibody (Ab) responses appear to be necessary for full demyelinating lesion to occur, both in experimental models and human MS. In the CSF, elevated levels of locally synthesized immunoglobulin (IG) and oligo-clonal Ab derived from clonally restricted plasma cells are also characteristic of MS. The pattern of oligoclonal banding is unique to each individual, and attempt to identify the target of these Abs have been largely unsuccessful. [19]

In this study we try to evaluate one of these auto antibodies which are MAG- Ab in the serum of patients with RRMS. In this evaluation we attempt to find any significant elevation of these auto antibodies in the serum of patients with RRMS). We found a higher percent of positive MAG antibodies among patients with RRMS (7.6%) than the control group (3.5%) but with no statistical significance as shown in table 2. This result is in agreement with two previous researches (Moller et al and Eduardo et al**)** **[17, 18]**. Failure of finding significant increase of anti-MAG in the serum of patients with RRMS may reflect low sized sample or not very sensitive test used.

Baig, et al. [20] demonstrated that antibodies secreting cells were less frequent in blood and bone marrow reflecting compartmentalization to CSF. This indicate that intrathecal production of anti- MAG, anti- MBP anti bodies may be important in the pathogenesis of MS. On the contrary Sato, et al [21] found positive anti MAG IgM in the serum of patients with demyelinating disease of the central and peripheral nervous system including: MS, sub-acute sclerosing pan encephalitis, Guillain Baree syndrome, chronic inflammatory demyelinating polyneuroppathy and others.[21]. lucchinetti, et al. reported four different pathologic subtypes of active MS lesion among 83 biopsies and autopsies of MS patients [4]. One of the subtypes, type 2 demonstrated a prominent presence of antibodies and complements. Another study [22] performed in 153 patients with acute, steroid-refractory CNS inflammatory disease demonstrated moderate to marked functional neurological improvement in 59% of the cases within 6 months following plasma exchange. The above studies indicating the importance of B cells and immunoglobulin in the pathogenesis of MS. Cepok et al. showed that B cells account for up to 25% of the CSF- infiltrating leukocyte during CNS inflammatory responses [23] .Another study showed strong correlation between level of oligoclonal band and prognosis for MS disability [24]. The above studies indicate the presence of autoantibody in the blood of patients with acute MS. The above results are compatible with our results in showing increase percent of positive anti MAG in the serum of patients with RRMS as indicated by pathological findings and response to plasma exchange therapy.

This study also showed that there is no clear impact of seropositive MAG- Abs on relapse of MS or disability as measured by EDSS tables (3, 4). The last two results were compatible with the previous studies [17-18]. This results may reflect the small sized sample or not very sensitive test used. Other important point in this study was the use of IgG rather than IgM, because IgG is highly associated with auto antibodies and indicate the chronic rather than the acute phase of the disease.

In conclusion there is a higher percent of anti MAG antibodies in the serum of patients with RRMS as compared with the control group. We recommend that these antibodies must be studied in the CSF of patients with RRMS because of compartmentalization and to choose the most sensitive assay because of its low concentration in the serum. Such futures studies that detect these Abs in the serum may be important in the diagnosis, treatment or prognosis.

Table 1. Distribution of study groups according to age (years).

Study group	**20-30**		**31-40**		**➢ 41**		Total	
	NO.	%	NO.	%	NO.	%	NO.	%
Multiple sclerosis	37	35.2	47	44.8	21	20	105	100
Control	28	32.9	30	35.2	27	31.8	85	100
Total	65	34.2	77	40.5	48	25.3	190	100

P value is >0.05, considered not significant.

Table 2. Distribution of IgG anti-myelin sheath auto-antibodies in study groups.

Study group	**Anti-myelin sheath Ab** (anti-MAG)*				**Total**	
	Positive		Negative			
	NO.	%	NO.	%	No	%
Multiple sclerosis	8	7.6	97	92.4	105	100
Control	3	3.5	82	96.5	85	100
Total	11	5.8	179	99.2	190	100

***P value is>0.05, considered not significant.**

Table 3. Distribution of IgG, anti-myelin sheath auto-antibodies, in study groups according to severity with EDSS.

Study group	**RRMS patient Anti-myelin sheath Ab** (anti-MAG)*				**Total**	
	Positive		Negative			
	NO.	%	NO.	%	No	%
With EDSS ≥ 3.5	2	6.2	30	93.8	32	100
With EDSS < 3.5	6	8.2	67	91.8	3	100
Total	8	7.6	97	92.4	105	100

***P value is>0.05, considered not significant.**

Table 4. Distribution of IgG, anti-myelin sheath auto-antibodies in RRMS study groups according to state at presentation (with relapse or without relapse).

Study group	**RRMS patient Anti-myelin sheath Ab** (anti-MAG)*				**Total**	
	Positive		Negative			
	NO.	%	NO.	%	No	%
With relapse	2	7.1	26	92.9	28	100
Without relapse	6	7.8	71	92.2	77	100
Total	8	7.6	97	92.4	105	100

***P value is>0.05, considered not significant.**

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