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Study of ABO System and Multiple Sclerosis disease in Iraq

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Abstract

Multiple sclerosis (MS) is one of an autoimmune condition with uncertain etiopathology. According to new data, ABO system had played a role in the development and understanding numerous diseases. Lower level of 25-hydroxy vitamin D3 (25-OHD3) is considered as a risk factor for MS. The aims of this study is to identify the role of blood group distribution on the levels of parathyroid hormone (PTH), 25-OHD3, total calcium, inorganic phosphorus and total magnesium on MS patients. Additionally, we assessed the relation between Expanded Disability Status Scale (EDSS) and study parameters in patients. The Study included 107 patients with MS were distributed in to four groups according to their blood group (A, B, AB, and O). Additionally, 124 apparently healthy individuals as control group. Tukey analysis was showed the level of 25-OHD3 in patients with B^+ was significant decrease than O^+ and A^+ patients group ($P \leq 0.05$). Furthermore, EDSS was negatively correlated with 25-OHD3 ($P \leq 0.05$) in B+ and O^+ patient groups. Through this study, ABO group may be consider as a risk factor for MS susceptibility as another interesting variable.

Keywords: Multiple sclerosis, ABO system, 25-OHD3, EDSS, Risk factor

دراسة نظام ABO ومرض تصلب الاعصاب المتعدد في العراق

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الخلاصة

تصلب الاعصاب المتعدد هو احد أمراض المناعة الذاتية الذي لا تزال أسبابه المرضية غيرمعروفة. تشير الأدلة الحديثة إلى أن نظام ABO قد يلعب دورًا في التسبب في العديد من الأمراض وفهمها. وكما يعد انخفاض مستوى25-هيدروكسي فيتامين د 3 (25-OHD3) احد عوامل الخطورة لمرض تصلب الاعصاب المتعدد، تهدف هذه الدراسة الى تحديد تركيز PTH و 25-OHD3 و الكالسيوم والمغنيسوم الكلي و والفوسفور اللاعضوي للمرضى ضمن فصائل الدم المختلفة.بالاضافة الى تقيم العلاقة بين ومقياس حالة الإعاقة الموسع و معايير الدراسة(EDSS) . اشتملت الدراسة على 107 مريضًا بالتصلب المتعدد تم توزيعهم على أربع مجاميع وفقًا لفصيلة الدم (A ، B، AB، O)علاوة على ذلك ، ضمت المجموعة الضابطة 124 فردًا سليمًا. أظهر التحليل الاحصائي Tukeyانخفاضًا معنويا ($P \leq 0.05$)

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لمستوى 25−OHD3 في مصل المرضى ضمن فصيلة الدم B^+ بالمقارنة مع المرضى الحاملين لفصيلة الدم O^+ و A^+ .علاوة على ذلك ، ارتبط EDSS سلبيا ولإحتمالية ($P\leq0.05$) بـ 25−OHD3 في مجموعات المرضى الحاملين لفصيلة الدم B^+ و O^+ .من خلال هذه الدراسة ، وبهذا يمكن تضمين مجاميع ABO كعامل خطورة مرتبط بمرض تصلب الاعصاب المتعدد كمتغير آخر مثير للاهتمام.

1. Introduction

In ABO system, blood groups are classified as dominant A and B and recessive O. The main genotypes that combine these three alleles are: A, B, AB and O. There are eight potential variations in which Rh status (positive or negative) and genotypes can be combined [1]. Although, both biological and clinical effects of blood groups have been discovered about a century ago, their full comprehension has yet to be reached. It has played a role not only with respect to haematology, transfusion and transplantation, but also in pathogenesis and awareness of a wide variety of diseases[2].

Blood groups specialize in the direct role of signal transducers, adhesion molecules, and pathogen receptors in the immune response to infections[1]. Some phenotypes were also linked to host resistance to specific pathogens, although their role in autoimmune conditions is uncertain [3]. Multiple sclerosis (MS) is one of an autoimmune disorder with an uncertain etiopathology. A number of studies have found a modest increase in MS susceptibility with gene-sensitivity and environmental factors especially (vitamin D3, Ultraviolet B exposure, infection by Epstein – Bar viruses, smoking and obesity) [4]. That induces immune responses to central nervous system auto-antigens [5]. MS has identified by Charcot in 1868 under the name of *"la sclerose en plaques"*. Charcot founded in his studies that MS lesions were named by *"plaques"* resulting from focal inflammation, demyelination, gliosis and various degrees of axonal loss [6]. MS is typically marked by dysfunction in a variety of neurological axes with numbness or paraesthesia, motor weakness, monocular visual disturbances, in coordination, dizziness, diplopia, sensory symptoms, bladder and sexual dysfunction, ataxia and vertigo[7].

Studies of 1980s and 1990s examined the relation between ABO and MS [8,9]. Several analytical epidemiological investigations have suggested an association between low levels of 25-hydroxyvitamin D3 (25-OHD3) and MS [10–13]. The biological origins of this relation are unclear. Hypotheses include mechanisms of cause, the genetic and environmental influences, or only the combination of various environmental factors. The relation between 25-OHD3 and inflammation risk was studied by most researchers [14], while others assessed the role of 25-OHD3 in myelination and re-myelination [15]. Thus, 25-OHvitD3 has vital roles in CNS functions, maturement of the immune cells, homeostasis of epithelial barrier and inhibiting proliferation and induces differentiation for different type of cells [16]. Also, the blood homeostasis for both calcium and phosphate is maintained via vitamin D with the PTH [17]

Typically, neurologists have assessed MS patient disability by "Expanded Disability Status Scale" (EDSS). John F. Kurtzke was first assigned the EDSS in 1983. The disability is calculated in eight functional systems: cerebellar, brainstem, pyramidal, sensory, visual, bowel and bladder, cerebral and others. The range of score values are from 1.0 to 10.0, with 10.0 reflecting to death as a result of MS [18,19]. Studies assessed the association between EDSS scores and serum 25-OHD3 level was considered as risk factors for MS progression [20].

The aims of this study are to identify the role of blood group distribution in MS patients and its effects on the serum levels of PTH, 25-OHD3, total calcium, inorganic phosphorus and total magnesium. Additionally, we assessed the relation between motor disability regarding to EDSS and study parameters concerning different blood groups of patients.

2 Experiment

2.1 Patients

The samples of current study were conducted from December 2019 to February 2020 at the Multiple Sclerosis Clinic/Medical City in Baghdad, Iraq. The study population included 107 MS patients with age range from 18 to 55 years as 50 male and 57 female, all patients were diagnosed according to McDonald criteria[21]. MS patients were distributed in to four groups according to their blood group ,group I involved 33 MS patients with blood group A^+, group II consisted 19 MS patients with blood group B^+, group III included 46MS patients with blood group O^+, and the group IV involved 9 MS patients with blood group AB^+. Additionally, 124 apparently healthy individuals (60male and 64 female) as control group with age range from 18 to 55 years were involved 56 with A^+, 36 with B^+, 18 with O^+ and 14 with AB^+ blood groups.

2.2 Samples collection

Five ml of blood was drawn from each individuals, and put in gel tubes at 25^{o}C for 15 minutes, at that time all tubes were centrifuged at 2000xg for 15 minutes. Finally, the sera was then stored at -40^{o}C until it was used.

2.3 Body Mass Index (BMI)

BMI was calculated according to below equation, obesity was defined as an BMI of 30 kg/m^2 or higher, overweight as an BMI 25.0 – 29.9, healthy weight was considered as an BMI =18.5 – 24.9, and underweight was defined when BMI is below 18.5 [21].

$$\text{BMI} = \text{Weight in (kg)}/\ \text{Height}^2 \text{ in } (\text{m}^2) \quad (1)$$

2.4 Laboratory analysis

Blood group for patients were identified with a Spinreact, Spain kit. Enzyme-Linked Immuno sorbent Assay (ELISA) kits from Mybiosource,USA were used to measure PTH and 25-OHD3 serum levels. While, BioSystems A15 analyzers were accomplished to measure total calcium, inorganic phosphorus and total magnesium serum levels.

2.5 Statistical analysis

The data of our study is presented as a mean ±standard deviation (mean± SD) and were analysed using SPSS statistics (version25). To assess the differences between groups, one-way variance analysis (ANOVA) was utilized. The significance level for the statistical analysis was P≤0.05.

3. Results

Table 1shows the demographics characteristics of MS patients and control groups. There were non-significant differences in age, weight, length, or BMI among the studied groups (P>0.05).

Table 1-Demographic characteristics of the patients and control groups.

Factors	Controls (N=124)	Patients (N=107)	P-Value
	mean± SD		
Age (years)	35.51±9.26	33.36 ±7.86	NS
Length (cm)	171.2±8.04	170.99±8.38	NS
Wight (kg)	71.50±10.09	72.14 ±11.75	NS
BMI (kg/m^2)	24.36±2.79	24.43±3.14	NS
Disease duration(years)	-	4.82±3.88	-
EDSS level	-	0.87±1.08	-

N: number of samples, SD: Standard division, NS: Non- significant P> 0.05.

Table 2 shows the mean ±SD value of the studied parameters in MS patients and the controls with group I and group II. Our results recognized that 25-OHD3 level was significantly decreased (P≤0.01) in patients than the control within group I (as 52.43 ± 17.54 ng/ ml vs 67.69±19.54 ng/ ml). Also, when compared patients within group I to the control group there was a significant increase (P≤0.05) in PTH levels (51.55±13.69 pg/mL vs 44.98±14.89 pg/ml) and a significant decrease (P≤0.05) in total calcium (8.42± 0.69 mg/dl vs 8.77±0.765 mg/dl). On the contrary, total magnesium level increased significantly (P≤0.01) in patients than

control group (2.03± 0.32 mg/dl vs 1.77 ±0.43 mg/dl).

Table 2- The serum level of the study parameters in MS patients and the control within group I and group II.(N: Number of samples, SD: Standard division, NS: Non- significant P> 0.05,* Significantly P≤0.05)

Parameters	Control/Control N=56/N=36	Group I/Group II N=33/N=19	P-value
	Mean ± SD		
25-OHD3 (ng/ ml)	67.69±19.54	52.43 ± 17.54	0.000*
	74.70±14.98	39.22±18.74	0.000*
PTH (pg/ml)	44.98±14.89	51.55±13.69	0.029*
	47.67±11.57	57.22±11.01	0.014*
Total calcium (mg/dl)	8.77±0.765	8.42± 0.69	0.022*
	8.78 ±0.66	7.99± 0.66	0.000*
Inorganic phosphorus (mg/dl)	2.85 ±0.57	2.90± 0.77	NS
	2.70±0.35	2.85± 0.35	NS
Total magnesium (mg/dl)	1.77 ±0.43	2.03± 0.32	0.001*
	1.69 ±0.32	1.95± 0.26	0.012*

As shown in Table 2 a significant decrease was observed in 25-OHD3 and total calcium serum levels in patients than the control with group II (P≤0.01) as (39.22±18.74 ng/ ml vs 74.70±14.98 ng/ ml) and (7.99± 0.66 mg/dl vs 8.78 ±0.66 mg/dl), respectively. In contrast in patients, there was an increase in PTH and total magnesium serum levels than in the control group with (P≤0.05) as (57.22±11.01 pg/mL vs 47.67±11.57 pg/ml) and (1.95± 0.26 mg/dl vs 1.69 ±0.32 mg/d), respectively. Statistical analysis in patients and the control within group III showed highly significant decrease (P≤0.01) in the 25-OHD3 level in patients than the control group (48.65±18.13 ng/ml vs 70.16±13.23 ng/ml). Similarly, in MS patients total calcium was significantly decreased (P≤0.05) as compared to the control group (8.20± 0.71 mg/dl vs 8.61±0.66 mg/dl); while there is a significant increase in patients serum level of total magnesium (P≤0.05) and inorganic phosphorus (P≤0.01) as compared to the controls groups of (1.96± 0.38 mg/dl vs 1.76±0.38 mg/dl) and (3.23± 0.86 mg/dl vs 2.45±0.64 mg/dl), respectively; while in group IV only 25-OHD3 was significantly decreased (P≤0.05) and the total magnesium was significantly increased (P≤0.05) in the patients than the controls with (45.85±16.79 ng/ml vs 68.27±10.22 ng/ml) and (.98± 0.13 mg/dl vs 1.52 ±0.30 mg/dl), see Table 3.

Table 3-The serum level of the studied parameters in patients and the control within group III and group IV. (N: Number of samples, SD: Standard division, NS: Non- significant P> 0.05,* Significantly P≤0.05)

Parameters	Control/Control N=18/ N=14	Group III/ Group IV N=46/ N=9	P-value
	Mean ± SD		
25-OHD3 (ng/ mL)	70.16±13.23	48.65±18.13	0.000*
	68.27±10.22	45.85±16.79	0.003*
PTH (pg/mL)	50.15±13.45	51.41± 14.41	NS
	46.90 ±15.06	53.11±9.76	NS
Total calcium (mg/dL)	8.61 ± 0.66	8.20± 0.71	0.013*
	8.97 ± 0.27	8.23± 0.75	NS
Inorganic Phosphorus (mg/dL)	2.45±0.64	3.23± 0.86	0.000*
	2.99±0.33	2.99± 0.57	NS
Total magnesium (mg/dL)	1.76±0.38	1.96± 0.38	0.047*
	1.52 ±0.30	1.98± 0.13	0.003*

As shown in Table 4A and B, there was a negative correlation (P≤0.05) between EDSS with 25-OHD3 in patients within Group II and Group III.

Table 4 A- EDSS correlation with other parameters in MS patients within group I and group II. (NS: Non- significant P> 0.05,* Significantly P≤0.05)

Component Vs EDSS	Group I N=33			Group II N=19		
	R^2	r	P	R^2	r	P
25-OHD3	0.147	-0.384	NS	0.244	-0.494*	0.032
PTH	0.015	0.121	NS	0.009	-0.097	NS
Total calcium	0.038	0. 195	NS	0.017	-0.131	NS
Inorganic phosphorous	0.102	-0.319	NS	0.020	-0.140	NS
Total magnesium	0.003	0.051	NS	0.006	-0.078	NS

Table 4 B-The EDSS correlation with other parameters in MS patients within group III and group IV. (NS: Non-significant P> 0.05,* Significantly P≤0.05).

Component Vs EDSS	Group III N=46			Group IV N= 9		
	R^2	r	P	R^2	r	P
25-OHD3	0.128	-0.358*	0.02	0.438	-0.66	NS
PTH	0.001	-0.032	NS	0.252	0.502	NS
Total calcium	0.003	0.055	NS	0.252	-0.500	NS
Inorganic phosphorous	0.004	-0.066	NS	0.070	0.264	NS
Total magnesium	0.002	0.049	NS	0.006	-0.080	NS

Tukey analysis in patients with different blood group showed a significant decrease in the serum level of 25-OHD3 in patients with B^+ than patients with A^+ and O^+ groups with (P≤0.01) and (P≤0.05), respectively, see Figure 1. Multiple comparisons analysis for patients with different blood group showed a significant decrease in the serum level of total calcium in patients with A^+ than patients with B^+ blood groups (P≤0.05), see Figure 2. Also, in O^+ patients group a significant increase was found in the level of inorganic phosphorus with (P≤0.05) than in patients with A^+ and B^+ groups Figure 3.

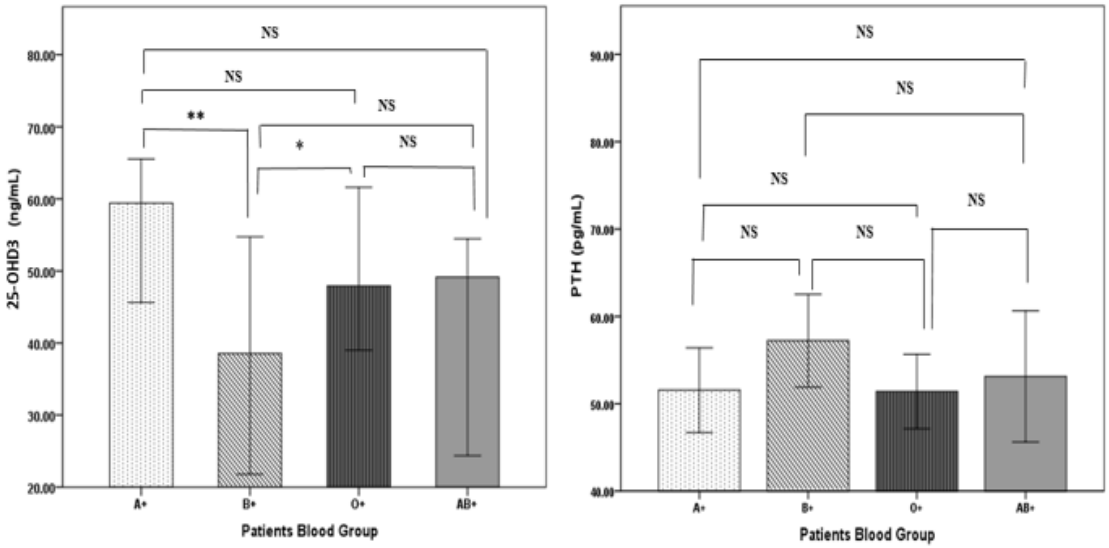

Figure 1-Tukey analysis for 25-OHD3 and PTH in MS patients with different blood groups. NS: Non-significant P> 0.05,* Significantly P≤0.05 and ** Significantly P≤ 0.01.

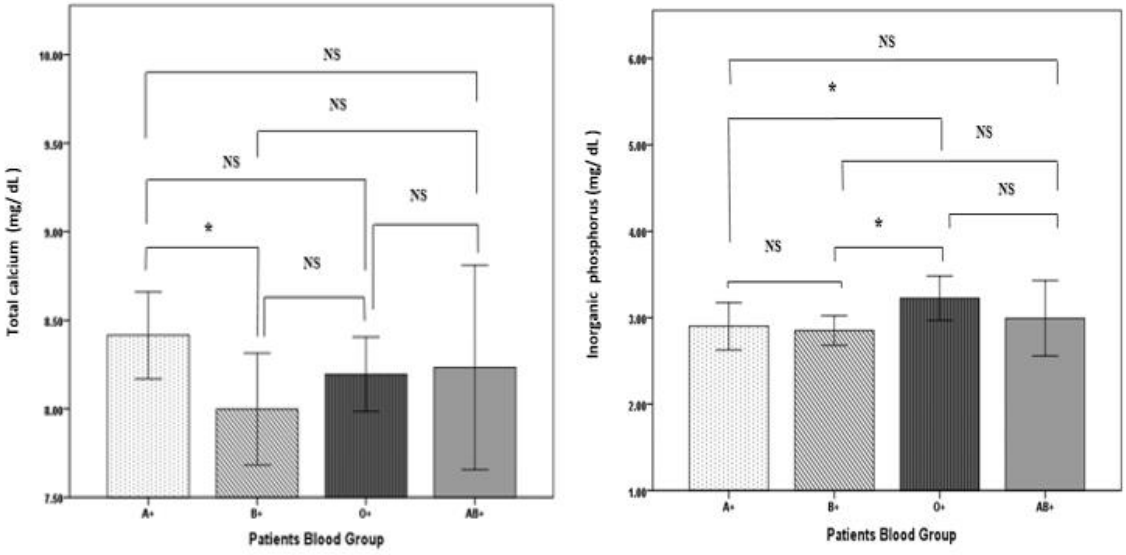

Figure 2-Tukey analysis for total calcium and inorganic phosphorus in MS patients with different blood groups. NS: Non-significant P> 0.05,* Significantly P≤0.05 and ** Significantly P≤ 0.01.

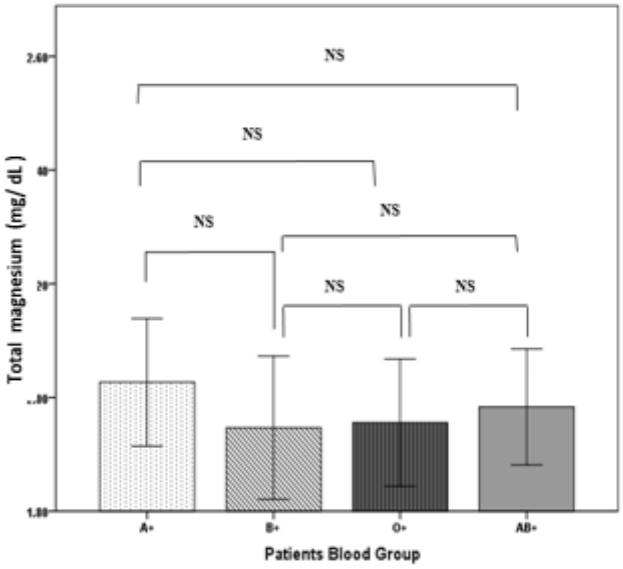

Figure 3- Tukey analysis for total magnesium in MS patients with different blood groups. NS: Non-significant P> 0.05,* Significantly P≤0.05 and ** Significantly P≤ 0.01.

4. Discussion

There is a significant evidence indicated that both the ABO system and a the low level of 25-OHD3 are relevant to MS pathogenesis. In this paper, in all patients, 25-OHD3 serum levels was significantly decreased than the healthy control groups. In MS patients, who have B^+ group, the lower serum level of 25-OHD3 was found as 39.222±18.743 (ng/ ml) than in other patient groups. Tukey analysis in patients with different blood group showed there was a significant decrease in the serum level of 25-OHD3 in patients with B^+ group than in O^+ and A^+ patients groups with (P≤0.05) and (P≤0.01), respectively. Furthermore, EDSS was negatively correlated with 25-OHD3 (P≤0.05) in B^+ and O^+ patients groups. In line with our results, multiple reports have investigated the links between the pathogenesis of MS and ABO groups; while another study findings were inconsistent, which found that A^+ and/or B^+ alleles

are a risk factor for MS [3]. Thus, several investigators suggested that a higher level of EDSS is correlated with lower 25-OHD3 serum level and with higher motor disability [15, 22]. As a result, lower 25-OHD3 serum levels contribute to irregular bone mineralization and hence to complications with movement. In view of the decreased probability of 25-OHD3 dermal synthesis in MS-patients with higher EDSS scores[23]. Association between 25-OHD3 and PTH with phosphorus and calcium are regulating the metabolism of calcium-phosphate [7]. Thus, PTH improves the reabsorption of calcium and enhances the production of 1,25-OHD3 and bone resorption. Furthermore, 1,25-OHD3 raises calcium and phosphate intestinal absorption. Deficiency of 25-OHD3 decreases intestinal absorption and secondary PTH secretion [24], which explained the higher level of PTH in MS patients with significantly decreased of 25-OHD3 and total calcium levels than control groups. This finding was confirmed by several studies [25, 26]. In addition, a substantial rise in total magnesium level in MS patients relative to control groups can be related to the daily use of magnesium supplementary by patients [27, 28].

5. Conclusion

Through this study, the results showed a significant relation between B^+ and O^+ blood groups with 25-OHD3 and EDSS which were considered as risk factors for MS disease. Therefore, ABO group may be considered as a risk factor for MS susceptibility as another remarkable variable. Our observations are confined to a small region. This association requires more studies in some other regions. Moreover, in further functional studies, the relevance of this interaction and its functional ramifications should be considered.

6. Acknowledgments

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7. Conflict of Interests: The authors declare that they have no conflicts of interest.

8. Ethics

This study was approved by Department of Chemistry, College of Science, Al-Mustansiriyah University, and Multiple Sclerosis Clinic / Medical-City in Baghdad, Iraq. Written consent was obtained from all MS patients who volunteered to participate in the study.

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