Current Journal of Neurology

Original Paper

Curr J Neurol 2021; 20(3): ?-?

Electrophysiological studies in patients with seropositive/seronegative myasthenia gravis

Received: 10 Mar. 2020
Accepted: 07 May 2021

Zahra Vahabi[1,2], Ferdos Nazari[3], Farzad Fatehi[3], Valiolah Bayegi[4,5], Zahra Saffarian[6], Fatemeh Saffarian[7], Shahriar Nafissi[3]

[1] Department of Geriatric Medicine, Ziaeian Hospital, Tehran University of Medical Sciences, Tehran, Iran
[2] Memory and Behavioral Neurology Division, Roozbeh Hospital, Tehran University of Medical Sciences, Tehran, Iran
[3] Department of Neurology, Shariati Hospital, Neuromuscular Research Center, Tehran University of Medical Sciences, Tehran, Iran
[4] Department of Epidemiology and Biostatistics, School of Public Health, Tehran University of Medical Sciences, Tehran, Iran
[5] Sina Trauma and Surgery Research Center, Tehran University of Medical Sciences, Tehran, Iran
[6] Department of Dermatology, School of Medicine, Imam Khomeini Hospital, Tehran University of Medical Sciences, Tehran, Iran
[7] Department of Cardiology, School of Medicine, Qazvin University of Medical Sciences, Qazvin, Iran

Keywords
Electrophysiology; Myasthenia Gravis; Patients

Abstract

Background: Myasthenia gravis (MG) affects the neuromuscular transmission, causing fluctuating muscle weakness and fatigue. This study is carried out with the aim to study the electrophysiologic findings of different subtypes of MG referred to our center in Tehran, Iran.

Methods: All patients with MG presenting to neurology department of Shariati Hospital, Tehran University of Medical Sciences were enrolled. Clinically, patients with MG were categorized as ocular vs. generalized. The acetylcholine receptor (Ach-R) and muscle-specific receptor tyrosine kinase (anti-MuSK) antibodies were performed. Repetitive Nerve Stimulation (RNS) was performed using the standard method, with supramaximal stimulation of muscles at the 3 Hz frequency by surface electrode at rest. Abductor pollicis brevis (APB) (median nerve), anconeus (radial nerve), trapezius (accessory nerve), and nasalis (facial nerve) muscles were studied in all patients. Single fiber electromyography (SFEMG) was performed by standard method.

Results: 196 seropositive patients with MG were included in the study. In electrophysiological studies, RNS was performed for 146 patients of Ach-R-Ab positive MG, with positive results in 110 patients. In addition, SFEMG was conducted for 8 patients with negative RNS, which resulted in 7 positive tests.

How to cite this article: Vahabi Z, Nazari F, Fatehi F, Bayegi V, Saffarian Z, Saffarian F, et al. Electrophysiological studies in patients with seropositive/seronegative myasthenia gravis. Curr J Neurol 2021; 20(3): ?-?.

Corresponding Author: Shahriar Nafissi
Email: nafisi@sina.tums.ac.ir

Among 23 patients with anti-MuSK-positive MG, RNS was performed for 16 patients, with positive results in 11 patients. The 5 remaining patients with negative RNS test were studied by SFEMG, 4 of whom had positive results. APB compound muscle action potential (CMAP) decrementation significantly correlated with Ach-R-Ab positive MG ($P < 0.03$).

Conclusion: This finding can support the hypothesis that the selection of muscles in electrodignostic study would be important. The electrodiagnostic studies are a good and non-invasive diagnostic tool for MG, and a combination of different distal, proximal, and facial muscles can increase the overall sensitivity of the test.

Introduction

Myasthenia gravis (MG) accounts for a heterogeneous group of autoimmune diseases, which affects the neuromuscular transmission, causing fluctuating muscle weakness and fatigue. Based on clinical course and muscle involvement, it can be categorized into ocular MG (about 20% of patients with MG) and generalized MG.[1]

The disease is caused by autoantibodies against post-synaptic components of neuromuscular junction. The most well-known autoantibodies include antibody against acetylcholine receptor (anti-Ach-R), antibody against muscle-specific receptor tyrosine kinase (anti-MuSK), and antibody against low-density lipoprotein related protein 4 (anti-LRP4). These antibodies are pathogenic factors and good diagnostic markers at the same time. Based on the presence or absence of these pathogenic factors, the disease can be categorized as anti-Ach-R-positive MG, anti-MuSK-positive MG, anti-LRP4-positive MG, and seronegative MG as well.[1,2] Given the literature, there are several clinical differences between the first two major types of seropositive MG. For instance, ocular myasthenia which is defined as restricted ocular muscle weakness after 2 years of disease onset, is rarely seen in anti-MuSK-positive MG, but it is not uncommon in anti-Ach-R-positive MG.[3]

This study is carried out to study the electrophysiologic findings of different subtypes of MG referred to our center in Tehran, Iran. To the best of our knowledge, there is no such study previously conducted in Iran.

Materials and Methods

The samples of the current study were selected using the combination of convenience and purposive sampling method among all eligible patients with MG referring to Neurology Department of Shariati Hospital, Tehran University of Medical Sciences. Diagnosis of MG was performed based on clinical findings (ocular, bulbar, and limb weakness and fatigue) at presentation, serum autoantibodies (AChR and MuSK), objective response to parenteral cholinesterase inhibitor (IV Edrophonium), and electrophysiological signs of abnormal neuromuscular transmission. The patients' data were used for the study after obtaining an informed consent form from them. The study inclusion criteria were all patients with MG who completed electrophysiologic investigation and also their serological profile was determined, and the exclusion criteria included patients with MG who lacked the desired document. This study was approved by the ethics committee of Tehran University of Medical Sciences.

Clinical Profile: Clinically, the patients with MG were categorized as ocular vs. generalized. The clinical characteristic of MG is fatigable weakness and ocular weakness is the most common initial presentation of MG, occurring in 85% of cases. MG weakness limited to ocular muscles after 2 years was considered as the ocular type. Clinical findings were classified as ocular, bulbar, cervical, limb, facial, axial, and respiratory weakness.

Antibody assay: The Ach-R antibodies were measured by a standard radioimmunoassay (RIA) method. Titers greater than 0.40 Nano-moles per liter were considered positive. Serological studies for autoantibodies against MuSK in seronegative patients were performed by Professor Angela Vincent's lab (Neurosciences Group, Weatherall Institute of Molecular Medicine, Oxford, UK). Due to some limitations, assay for the Anti-MuSK antibody was performed on only 41 seronegative patients. Out of them, 23 patients were anti-MuSK-positive, who were enrolled in the study.

Electrophysiological assessment: Repetitive Nerve Stimulation (RNS) was performed using the standard method, with supramaximal stimulation of muscles at a frequency of 3 Hz by surface electrode at rest. The Abductor pollicis brevis (APB) (median nerve), anconeus (radial nerve), trapezius (accessory nerve), and nasalis (facial nerve) muscles were studied in all patients. Abnormal result was defined as equal or greater than 10% decrement of the compound muscle action potential (CMAP) amplitude of the fourth response compared to the first.[4] If RNS was negative at rest, exercise test was used. The muscle

was exercised for one minute and then stimulated immediately, and after 1, 2, 3, and 4 minutes, the abnormal result was again defined as equal or greater than 10% decrement of the CMAP amplitude of the fourth response compared to the first again. Single fiber electromyography (SFEMG) was performed by the standard method and abnormal SFEMG study was defined as having increased jitter in more than 10% of the potential pairs measured, or a mean jitter value exceeding the upper limit of the normal for the examined muscle.[5] SFEMG was performed when RNS test was negative and facial muscles (e.g., orbicularis oculi and facialis) were preferred.

The baseline characteristics were presented in the form of numbers (%) or mean [with standard deviation (SD)]. Besides, the chi square test and Mann-Whitney U-test were applied for comparison of clinical features between the anti-Ach-R-positive MG and anti-MuSK-positive MG and mean decrement in the electrophysiological features, respectively. Statistical significance was defined as $P < 0.05$ and the statistical analyses were performed with STATA software version 14.2.

Results

After data collection, 196 seropositive patients with MG were included. The mean age of the Ach-R-Ab positive and MuSK-Ab positive patients with MG was 36.0 ± 15.9 and 35.5 ± 15.4, respectively. There was no significant difference in age of the two groups of patients ($P = 0.88$), as well as in the clinical profile of the two groups (Table 1).

In electrophysiological studies, RNS was performed for 146 patients of Ach-R-Ab positive MG, with positive results in 110 patients. Moreover, SFEMG was conducted for 8 patients with negative RNS, which resulted in 7 positive tests. Among 23 patients with anti-MuSK-positive MG, RNS was performed for 16 patients, with positive results in 11 patients. The 5 remaining patients with negative RNS test were studied by SFEMG, 4 of whom had positive results. The details of these studies are reported in table 2.

Discussion

MG is an autoimmune disease with fluctuating fatigue and muscle weakness. Weakness usually involves extraocular, bulbar, limb, and axial muscles. Specifically, 60 of anti-Ach-R positive patients with MG and about one-third of anti-MuSK-positive patients with MG present with ptosis and diplopia.[2] Nevertheless, in our series, 71 of anti-Ach-R positive patients with MG and 73 of anti-MuSK positive patients with MG had ocular presentations at the baseline. There are several important clinical differences between these two types of MG. For example, more than 40 of anti-MuSK-Ab positive patients present with bulbar symptoms, usually coupled with neck and respiratory muscle weakness.[2] In a series of 53 MuSK-Ab positive patients from the United States, about three quarters of the patients had bulbar involvement.[6] Although we encountered common bulbar involvement at presentation (56) in our anti-MuSK-positive patients with MG, neck or respiratory muscle weakness was not common as presenting symptoms. Another interesting finding in our series was more common ocular MG and ocular findings at presentation in the MuSK-Ab-positive patients, which is inconsistent with the studies above, for which we have no explanation.

One of the widely used diagnostic tools for MG is the RNS test. During this test, muscle response (CMAP) to surface nerve stimulation is recorded. Indeed, the decrementing CMAPs amplitude in response to RNS is a sensitive tool for MG diagnosis.[7]

Table 1. Baseline characteristics of participants as mean ± standard deviation (SD) or n (%)

Variable		Ach-R-Ab positive (n = 173)	MuSK-Ab positive (n = 23)	P
Age (year) (mean ± SD)		36.0 ± 15.9	35.5 ± 15.4	0.88
Sex [n (%)]	Male	65 (37)	7 (30)	0.51
	Female	108 (63)	16 (70)	0.51
Type [n (%)]	Ocular	21 (13)	4 (17)	0.59
	Generalized	152 (87)	19 (83)	0.59
Involved muscles at	Ocular	124 (71)	17 (73)	0.84
presentation [n (%)]	Respiratory	15 (8)	3 (13)	0.42
	Bulbar	88 (51)	13 (56)	0.65
	Limb	80 (46)	6 (26)	0.69
	Cervical	14 (8)	2 (8)	> 0.99
	Facial	35 (20)	4 (17)	0.73

SD: Standard deviation; Ach-R-Ab: Acetylcholine receptor antibody; MuSK-Ab: Muscle-specific receptor tyrosine kinase antibody

Table 2. Comparison of the two groups in the study variables

Procedure		Ach-R-Ab positive	MUSK-Ab positive	P
RNS	Negative	36 (24.7)	5 (31.2)	
	Positive	110 (75.3)	11 (68.8)	0.57
	n	146	16	
SFEMG	Negative	1 (12.5)	1 (20.0)	
	Positive	7 (87.5)	4 (80.0)	0.72
	n	8	5	
APB	Negative	41 (41.0)	7 (77.8)	
	Positive	59 (59.0)	2 (22.2)	0.03
	n	100	9	
ANC decrement	Negative	39 (50.6)	6 (75.0)	
	Positive	38 (49.4)	2 (25.0)	0.19
	n	77	8	
TRAP decrement	Negative	70 (58.8)	6 (50.0)	
	Positive	49 (41.2)	6 (50.0)	0.68
	n	119	112	
Nasalis decrement	Negative	29 (31.5)	2 (25.0)	
	Positive	63 (68.5)	6 (75.0)	0.70
	n	92	8	
APB Decrement Positive		27.53 (15.7)	22.50 (9.2)	0.65
	n	59	2	
ANC decrement Positive		25.91 (15.8)	20.00 (5.6)	0.60
	n	38	2	
TRAP Decrement Positive		27.51 (15.9)	20.50 (9.2)	0.29
	n	49	6	
Nasalis Decrement Positive		33.27 (18.6)	18.67 (4.5)	0.06
	n	63	6	

RNS: Repetitive Nerve Stimulation; Ach-R-Ab: Acetylcholine receptor antibody; MuSK-Ab: Muscle-specific receptor tyrosine kinase antibody; SFEMG: Single fiber electromyography; APB: Abductor pollicis brevis; ANC: Anconeus; TRAP: Trapezius

In a prospective study by Bou Ali et al., the global sensitivity of the RNS test was 82 and its specificity was 100.[8] Some studies suggested relationships between the degree of the CMAP amplitude decrement in different muscle groups and disease severity.[9] Another test with greater sensitivity is SFEMG, which uses a fine needle to record the conduction time and its variability in a single muscle fiber.[7] In a series of 486 patients with MG from India, 82.35 of the patients with generalized myasthenia irrespective of their antibody situation, had positive RNS test.[10] In another series of MuSK-Ab-positive patients, positive RNS test was reported in 83 of the patients, and positive SFEMG was observed in 90 of them.[6] Authors of a prospective study on 112 patients with MG from Thailand reported abnormal RNS test in 62 of ocular and 80 of generalized patients with MG. They also found positive SFEMG in 93 of ocular and 99 of generalized patients with MG, respectively.[11] A prospective study of 31 MuSK-Ab-positive and 28 Ach-R-Ab-positive patients from Serbia recorded positive RNS and positive SFEMG in 51.6 and 90.3 of MuSK-Ab-positive and 92.9 and 92.9 of Ach-R-Ab-positive patients, respectively.[12] In a series of 45 MuSK-Ab-positive patients with MG from Korea, abnormal RNS responses in limb and facial muscles were recorded in 22.2 and 77.8 of patients, respectively.[13] Authors of a retrospective study from Greece observed abnormal RNS test in 82.6 of patients with MG, with orbicularis oculi muscle considered as the most sensitive muscle for RNS test.[14]

The findings of our study are congruent with the results of other studies; we found positive RNS in 75.5 of Ach-R-Ab-positive and 68 of MuSK-Ab-positive patients. Our SFEMG results were also consistent with other studies with 87 and 80 positivity, respectively. There were no significant differences between the positive results of the two groups. The differential decrements of various groups of muscles (Anconeus, Trapezius, Nasalis) in our study showed no significant differences either. Only the APB CMAP decrementation significantly correlated with Ach-R-Ab positive MG ($P < 0.03$). This finding can support our

hypothesis that the selection of muscles in electrodignostic study would be important. The electrodiagnostic studies are a good and non-invasive diagnostic tool for MG, and a combination of different distal, proximal, and facial muscles can increase the overall sensitivity of the test.

Conclusion

This was a retrospective study conducted on seropositive patients with MG either with Ach-R or MuSK antibodies. Although our MuSK-Ab-positive patients were far fewer than the Ach-R-Ab-positive patients, there was a significant difference in the electrophysiological findings between the two groups. Further studies with a larger number of patients are required to confirm the results of the current study.

Limitations: One of the limitations of the current study was due to the cross-sectional methodological perspective used to answer the study questions, which made it difficult to reach a causal conclusion. The authors tried to minimize the effect of this limitation by grouping after measurement and extracting appropriate statistical indicators. We recommend a case-referent method for future studies.

Conflict of Interests

The authors declare no conflict of interest in this study.

Acknowledgments

We greatly appreciate the kindness and generosity of all patients for participating in this study.

References

1. Sieb JP. Myasthenia gravis: An update for the clinician. Clin Exp Immunol 2014; 175(3): 408-18.
2. Gilhus NE, Verschuuren JJ. Myasthenia gravis: Subgroup classification and therapeutic strategies. Lancet Neurol 2015; 14(10): 1023-36.
3. Jing F, Cui F, Chen Z, Yang F, Ling L, Huang X. Clinical and electrophysiological markers in myasthenia gravis patients. Eur Neurol 2015; 74(1-2): 22-7.
4. Abraham A, Alabdali M, Alsulaiman A, Breiner A, Barnett C, Katzberg HD, et al. Repetitive nerve stimulation cutoff values for the diagnosis of myasthenia gravis. Muscle Nerve 2017; 55(2): 166-70.
5. Kouyoumdjian JA, Stalberg EV. Reference jitter values for concentric needle electrodes in voluntarily activated extensor digitorum communis and orbicularis oculi muscles. Muscle Nerve 2008; 37(6): 694-9.
6. Pasnoor M, Wolfe GI, Nations S, Trivedi J, Barohn RJ, Herbelin L, et al. Clinical findings in MuSK-antibody positive myasthenia gravis: A U.S. experience. Muscle Nerve 2010; 41(3): 370-4.
7. Liang T, Boulos MI, Murray BJ, Krishnan S, Katzberg H, Umapathy K. Detection of myasthenia gravis using electrooculography signals. Annu Int Conf IEEE Eng Med Biol Soc 2016; 2016: 896-9.
8. Bou Ali H., Salort-Campana E, Grapperon AM, Gallard J, Franques J, Sevy A, et al. New strategy for improving the diagnostic sensitivity of repetitive nerve stimulation in myasthenia gravis. Muscle Nerve 2017; 55(4): 532-8.
9. Yan C, Song J, Pang S, Yi F, Xi J, Zhou L, et al. Palpebral portion of the orbicularis oculi muscle to repetitive nerve stimulation testing: A potential assessment indicator in patients with generalized myasthenia gravis. J Clin Neurosci 2018; 48: 238-42.
10. Patil SA, Bokoliya SC, Nagappa M, Taly AB. Diagnosis of myasthenia gravis: Comparison of anti-nicotinic acetyl choline receptor antibodies, repetitive nerve stimulation and Neostigmine tests at a tertiary neuro care centre in India, a ten-year study. J Neuroimmunol 2016; 292: 81-4.
11. Witoonpanich R, Dejthevaporn C, Sriphrapradang A, Pulkes T. Electrophysiological and immunological study in myasthenia gravis: Diagnostic sensitivity and correlation. Clin Neurophysiol 2011; 122(9): 1873-7.
12. Nikolic A, Basta I, Stojanovic VR, Stevic Z, Lavrnic D. Electrophysiological profile of the patients with MuSK positive myasthenia gravis. Neurol Res 2014; 36(11): 945-9.
13. Kim SW, Sunwoo MK, Kim SM, Shin HY, Sunwoo IN. Repetitive nerve stimulation in musk-antibody-positive myasthenia gravis. J Clin Neurol 2017; 13(3): 287-92.
14. Zambelis T, Kokotis P, Karandreas N. Repetitive nerve stimulation of facial and hypothenar muscles: Relative sensitivity in different myasthenia gravis subgroups. Eur Neurol 2011; 65(4): 203-7.