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Unveiling Myasthenia Gravis: A Comprehensive Analysis of Diagnostic Tools and Clinical Insights

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Abstract

Objective: This research offers a comprehensive analysis of Myasthenia Gravis (MG), uncovering the remarkable accuracy of spinal accessory, ulnar, and facial nerve repetitive nerve stimulation (RNS), along with the precision of single fiber electromyography (SF-EMG) in MG diagnosis. We also embark on an exploration of clinical features and autoantibody test results in generalized MG patients.

Methods: In this prospective study, we welcomed 31 individuals definitively diagnosed with generalized MG into our quest. The categorization of patients was conducted in accordance with the criteria set by the Myasthenia Gravis Foundation of America (MGFA). We examined patients' trapezius, nasalis, and abductor digiti minimi (ADM) muscles using RNS. We meticulously recorded the presence of MG autoantibodies, clinical subtypes based on affected muscle groups, and SF-EMG jitter rates.

Results: The mean age of the 31 patients of whom 19 (61.3%) were male, was 64 ± 13.9 years. Among them, 20 showed positivity in the Anti-AChR antibody test. In 28 patients, accounting for 90.3% of the study group, single fiber electromyography (EMG) displayed increased jitter. There were 4 (12.9%), 24 (77.4%) and 12 (38.7%) patients featuring a decremental response of exceeding 10% in ADM, trapezius and nasalis muscles, respectivelyOur investigation revealed notable findings, such as the absence of substantial correlations between decremental response rates and age, gender, duration of complaints, antibody test results, thymus abnormalities, affected muscle types, familial history, or increased jitter rates in SF-EMG ($p>0.05$).

Conclusion: As our findings clearly show, we can confidently attest to the remarkable sensitivity of RNS in MG diagnosis when muscle selection is precise. A gem discovered on our study is the high sensitivity of the spinal accessory nerve, a revelation that should guide the course of routine RNS studies, particularly for those facing ocular-onset myasthenia.

Keywords: Generalized Myasthenia Gravis, Trapezius Muscle, Repetitive nerve stimulation, Decrement

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Miyastenia Gravis'te Diagnostik Araçların ve Klinik Bulguların Kapsamlı Bir Analizi

Öz

Amaç: Bu çalışma, Miyastenia Gravis'e (MG) derinlemesine bir bakış sunarak, spinal aksesuar, ulnar ve fasiyal sinir ardışık sinir uyarımı (ASU) ile tek lif elektromiyografinin (SF-EMG) MG teşhisindeki duyarlılığını ortaya çıkarmaktadır. Ayrıca, jeneralize MG hastalarında klinik özellikler ve otoantikor test sonuçları değerlendirilmektedir.

Yöntemler: Bu prospektif çalışmaya, jeneralize MG tanısı kesin olarak konmuş 31 hasta dahil edilmiştir. Hastaların sınıflandırılmasında Miyastenia Gravis Amerika Vakfı (MGFA) kriterleri kullanılmıştır. Trapezius, nasalis ve abdüktör digiti minimi (ADM) kasları ASU kullanılarak incelenmiş, MG otoantikorlarının varlığı, etkilenen kas gruplarına göre klinik alt tipleri ve SF-EMG jitter oranları kaydedilmiştir.

Bulgular: 19'u (%61,3) erkek olan 31 hastanın yaş ortalaması 64 ± 13.9 yıldı. Bu hastalardan 20'sinde Anti-AChR antikor testinde pozitiflik gözlendi. Tek lif EMG, 28 (%90,3) hastada artmış jitter gösterdi. ADM, trapezius ve nasalis kaslarında %10'un üzerinde dekrement gösteren sırasıyla 4 (%12,9), 24 (%77,4) ve 12 (%38,7) hasta vardı. Araştırmamızda, dekrement oranları ile yaş, cinsiyet, şikayet süresi, antikor test sonuçları, timus anormallikleri, etkilenen kas tipleri, aile geçmişi veya SF-EMG'deki artmış jitter oranları arasında anlamlı korelasyon saptanmadı ($p>0.05$).

Sonuç: Bulgularımız, kas seçimi hassas olduğunda ASU'nun MG teşhisinde dikkat çekici duyarlılığını göstermektedir. Yüksek duyarlılığı göz önüne alındığında, spinal aksesuar sinir, özellikle oküler başlangıçlı miyasteni durumunda rutin RNS çalışmalarına dahil edilmelidir.

Anahtar kelimeler: Jeneralize Miyastenia Gravis, Ardılık sinir uyarımı, Trapezius Kası, Dekrement.

INTRODUCTION

Myasthenia Gravis (MG) is identified as an autoimmune disorder affecting the neuromuscular junction, leading to muscle weakness in the ocular, bulbar, and limb regions, where neuromuscular transmission is impeded[1]. The symptoms of MG, which progresses with remissions and relapses, typically fluctuate during the day. Respiratory muscle involvement can be observed in 15-20% of the MG patients. The etiology of MG has not been fully clarified. However, the role of thymus pathologies in the etiology of MG has been emphasized in the literature[2]. The diagnosis of MG is made based on the presence of clinical findings, positive response to anticholinesterases, serum anti-acetylcholine receptor antibody (anti-AChR) or antibody against muscle-specific kinase (anti-MuSK) positivity, and the results of the electrophysiological tests, e.g., repetitive nerve stimulation (RNS) and single fiber electromyography (SF-EMG)[3,4].

Given its non-invasive nature and ease of access, Repetitive Nerve Stimulation (RNS) is extensively employed in diagnosing conditions that impact the neuromuscular junction, including MG[3]. The purpose of RNS is to reduce presynaptic acetylcholine release with each low frequency stimulation. In a normal muscle, no reduction is expected in compound muscle action potential (CMAP) amplitude as a result of RNS often at a frequency of 3 Hz. A decrement is described as a decrease in the CMAP amplitude of the 4th stimulus by more than 10% compared to the 1st stimulus as a result of RNS performed in a symptomatic muscle[4]. The most commonly examined muscles by RNS are nasalis, orbicularis oculi, trapezius, abductor digiti minimi (ADM), deltoid and flexor carpi radialis (FCR) muscles5. The diagnostic accuracy of RNS increases in the event of proximal and facial muscles, and also fluctuates depending on the disease's severity. Reports indicate that the sensitivity of RNS ranges from 30% to 80% in the event of distal muscles and diseases with a mild course, and even lower in the event of ocular myasthenia[5,6].

Therefore, SF-EMG stands out as the most sensitive technique for detecting a defect in the neuromuscular junction[7,8]. Jitter refers to the variation in time intervals between action potentials from two distinct muscle fibers innervated by the same motor neuron, across successive firings of that neuron. A delay exceeding a certain period between the action potentials of the said two muscle fibers indicates a high jitter value, which, in turn, implies pathology in the motor endplate[7,9]. The sensitivity and specificity values of SF-EMG were found to be high in different studies, particularly in the event of generalized myasthenia[10,11]. According to the diagnostic algorithm of MG, first, antibody test is performed, followed by RNS, if antibody test positivity is not detected, and then SF-EMG, if no response is obtained in RNS. RNS techniques are widely used since performance of SF-EMG poses some technical difficulties. However, they must be performed with due care and from the most sensitive muscle for best results. Although RNS can be performed from different muscles in practice, it reportedly has higher sensitivity when performed from the proximal muscles rather than the distal muscles[4,12].

Considering the aforementioned context, this study aims to evaluate and contrast the accuracies of spinal accessory, ulnar and facial nerve RNS and SF-EMG in the diagnosis of MG and to investigate the clinical features and autoantibody test results in patients with generalized MG.

METHODS

The sample of this prospective study consisted of 31 patients who applied to Sakarya University Education and Research Hospital, Department of Neurology and were diagnosed with generalized MG. The definitive diagnosis of MG was made based on the presence of at least two of the relevant criteria concerning immunological findings, electrodiagnostic tests, and pharmacological response, in addition to typical clinical findings. Participants were included if they were over 18 years of age, had a definitive diagnosis of MG with complete antibody tests and electrodiagnostic tests, and were not on any medication that could disrupt neuromuscular transmission during the tests. Included patients had not taken pyridostigmine within the last 12 hours before the EMG test. Any patient who was recommended for a diagnostic assessment of a disorder related to neuromuscular transmission, identified through noticeable muscle fatigue, and exhibited significant decrement (10% or more) or showed atypical findings in single-fiber electromyography (SFEMG) during the electrodiagnostic examination, was considered a suitable candidate. Patients for whom electrophysiological examinations were contraindicated (such as those with bleeding diathesis), those with a malignancy or any other disease that may result in neuromuscular junction disease, and those with any condition that could impact the test results were excluded from the study.

The research methodology received authorization from the institution's review board (12.09.22/274). This study was conducted in strict compliance with the ethical standards set by the IJSM and adhered to the principles delineated in the Declaration of Helsinki. Patients for whom electrophysiological examinations were contraindicated (bleeding diathesis, etc.) and those with any disease that could impact the test results were excluded from the study. All patients underwent a comprehensive neurological examination, including a detailed examination of muscle strength. Patients' data including age, gender, detailed clinical history, presence of Anti-AChR antibodies and Anti-MuSK antibodies were collected and evaluated.

Patients were categorized according to the Myasthenia Gravis Foundation of America (MGFA) classification. The categorization of

clinical MG subtypes, as defined by the MGFA classification, was determined from the neurological records of patients or the notes of the referring doctors, typically 1 to 3 weeks prior to conducting RNS. Patients with mild non-ocular muscle weakness, including any level of ocular muscle weakness, were classified under MGFA class II. Those with moderate non-ocular muscle weakness, regardless of the severity of ocular muscle weakness, fell into MGFA class III. Lastly, individuals exhibiting severe non-ocular muscle weakness, along with ocular muscle weakness of any degree, were placed in MGFA class IV[13].

In the nerve conduction studies, we stimulated specific nerves at various locations: the facial nerve was stimulated at the mandibular joint, the accessory nerve at the neck, and the ulnar nerve at the wrist. This was done to capture the unilateral compound muscle action potentials (CMAPs) of the nasalis, trapezius, and abductor digiti minimi (ADM) muscles, respectively. RNS was performed in these muscles while at rest. Recordings were performed with a Key-point Classic EMG device (Alpine Biomed, Skovlunde, Denmark) and standard surface electrodes with a 10 mm diameter were used. Ten stimulations were performed on each muscle while at rest using supramaximal stimulation (3 Hz). The cut-off limit for neuromuscular conduction failure was accepted as a reproducible 10% reduction in peak amplitude from baseline to negative by comparison of first CMAP to fourth CMAP, consistent with previous analyses. The examination was conducted thrice, with a minimum interval of five minutes between each repetition. Any decrement studies deemed of subpar technical quality were omitted from subsequent analysis.

Subsequently, single fiber EMG examination was performed in all patients. SF-EMG data obtained from these recordings were indicated as abnormal, (i.e., increased mean consecutive difference for all pairs or increased jitter in 2 of 20 fiber pairs) or normal. All electrophysiological studies were performed at appropriate temperature in patients who were not on any medication or at least 4 to 12 hours after the last acetylcholinesterase inhibitor drug use in patients who were on medication. In the upper extremity nerve examinations, the immobilization of the extremity was achieved with small sandbags.

Age, gender, duration of complaints, presence of anti-AChR or anti-MuSK antibodies, clinical subtype according to the affected muscle group, presence of thymoma, and familial features were recorded. Significant decrement rates were compared with clinical and demographic data and SF-EMG results.

Statistical Analysis

The statistical analysis for this research was conducted using the R-Studio software. For quantitative variables, descriptive statistics were presented including the mean, standard deviation, median, and the minimum and maximum values; whereas qualitative variables were represented through their frequency and percentage. The Shapiro Wilk test was applied to determine if the quantitative variables conformed to a normal distribution. For comparing two independent groups of quantitative variables that deviated from normal distribution, the Mann-Whitney U test was employed. The Fisher-Freeman-Halton chi-square test, Fisher's chi-square test, and Yates chi-square test were utilized for the comparative analysis of qualitative variables between groups. In the statistical evaluation of this study, results that yielded a p-value below 0.05 were considered statistically significant.

RESULTS

The mean age of the 31 patients, 19 (61.3%) were male and 12 (38.7%) were female, was 64 ± 13.9 (min. 24, max. 80) years. In terms of MGFA subgroups, 7 patients were evaluated as class II, 14 were class III, and 10 were class IV. Anti-AChR

antibody test result was positive in 20 patients, anti-MuSK antibody test result was positive in 2 patients, and 9 patients were seronegative.

Analysis of the patients according to clinical symptomatology revealed ptosis, diplopia, bulbar muscle involvement and neck muscle involvement in 25 (80.6%), 10 (32.2%), 15 (48.3%), and 18 (58%) patients, respectively. The mean MG composite score was calculated as 3.09 ± 0.74. Thymoma was detected in four (12.9%) and thymic hyperplasia was detected in in one patient. Familial history was present in two (6.4%) patients. Single fiber EMG revealed increased jitter in 28 (90.3%) patients (Table I).

Table I: Patients' characteristics (n = 31)

		n	%
Gender	**Female**	12	38.7%
	Male	19	61.3%
Antibody test result	**AChR-Ab+**	20	64.5
	MuSK-Ab+	2	6.4
	Seronegative	9	29
Clinical findings	**Ptosis**	25	80.6
	Diplopia	10	32.2
	Bulbar	15	48.3
	Neck	18	58
Thymic abnormalities	**Thymoma**	4	12.9
	Thymic hyperplasia	1	3.2
	Normal	26	83.8
Positive Decrement (>10%)	**Ulnar**	4	12.9
	Trapez	24	77.4
	Nasalis	12	38.7
SF-EMG	**Abnormal Jitter**	28	90.3
	Normal Jitter	3	9.7
Familial MG	**Yes**	2	6.4
	No	29	90,9
MGFA classification	**Class II**	7	12.9
	Class III	14	3.2
	Class IV	10	83.8

Abbreviations: MG, Myasthenia Gravis; AChR-Ab, acetylcholine receptor antibody; MGFA, Myasthenia Gravis Foundation of America; MuSK-Ab, muscle-specific tyrosine kinase antibody; SF-EMG, Single Fibre Electromyography

In repetitive nerve stimulation, there were four (12.9%), 24 (77.4%) and 12 (38.7%) patients featuring a decremental response of above 10% in ADM, trapezius and nasalis muscles, respectively. No substantial correlation was observed between the rates of decremental response in all three muscles and age, gender, duration of complaints, antibody test positivity, presence of thymus abnormality, affected muscle type, family history, and increased jitter rates in SF-EMG ($p>0.05$) (Table II for trapezius muscle).

Table II: The relationship between presence of relevant decrement rate over trapezius muscle and MG characteristics

		Relevant decrement rate over trapezius muscle (n=31) Negative (n=7)	Positive (n=24)	p-value
Mean age (years)		61.43 ± 15.40	60.92 ± 13.08	0.695*
Symptom duration (months)		23.43 ± 31.16	13.31 ± 19.41	0.800*
Gender	**Female**	4 (57.1%)	8 (33.3%)	0.384**
	Male	3 (42.9%)	16 (66.7%)	
Antibody test result	**AChR-Ab+**	5 (71.4%)	15 (62.5%)	0.736+
	MuSK-Ab+	0 (0%)	1 (4.2%)	
	Seronegative	2 (28.6%)	8 (33.3%)	
Ptosis	**Absent**	2 (28.6%)	4 (16.7%)	0.596**
	Present	5 (71.4%)	20 (83.3%)	
Diplopia	**Absent**	6 (85.7%)	17 (70.8%)	0.642**
	Present	1 (14.3%)	7 (29.2%)	
Bulbar	**Absent**	3 (42.9%)	13 (54.2%)	0.685**
	Present	4 (57.1%)	11 (45.8%)	
Neck	**Absent**	4 (57.1%)	9 (37.5%)	0.413**
	Present	3 (42.9%)	15 (62.5%)	
Thymic abnormalities	**Thymoma**	0 (0%)	4 (16.7%)	0.522+
	Thymic hyperplasia	1 (14.3%)	0 (0%)	
	Normal	6 (85.7%)	20 (83.3%)	
SF-EMG	**Abnormal Jitter**	6 (85.7%)	22 (91.7%)	0.550**
	Normal Jitter	1 (14.3%)	2 (8.3%)	
Familial MG	**Yes**	0 (0%)	3 (12.5%)	1.00**
	No	7 (100%)	21 (87.5%)	
MGFA	**Class II**	2 (28.6%)	5 (20.8%)	0.546+
	Class III	4 (57.1%)	10 (41.7%)	
	Class IV	1 (14.3%)	9 (37.5%)	

**Mann-Whitney U test; **Fisher's Chi-Square test; +Fisher-Freeman-Halton Chi-Square test*

Abbreviations: MG, Myasthenia Gravis; MGFA, Myasthenia Gravis Foundation of America; AChR-Ab, acetylcholine receptor antibody; MuSK-Ab, muscle-specific tyrosine kinase antibody; SF-EMG, Single Fibre Electromyography

DISCUSSION

The findings of this study revealed that spinal accessory nerve stimulation is the most sensitive technique for RNS after SF-EMG in patients with generalized MG. On the other hand, there was no significant correlation between the decrement rates in RNS measurements measured by all three recording methods and clinical subtypes of the disease or autoantibody test results.

Varying sensitivity rates of RNS have been documented in scholarly works, contingent on the muscle type[12,14-16]. In one of these studies, Costa et al. stated that the trapezius, one of the shoulder muscles, should definitely be studied in neck-extremity-onset myasthenia, and recommended the use of the anconeus-nasalis muscles in bulbar myasthenia. In the event of ocular myasthenia, on the other hand, they reported that RNS was not sufficient and jitter increase should be sought with SF-EMG[14]. In another study, diagnostic sensitivity of RNS was calculated as 68.8%, 53.3%, 50.0%, 30.0%, and 16.7%, in the nasalis, trapezius, occipitalis, abductor pollicis brevis (APB), and ADM muscles, respectively. In addition, it has been reported that diagnostic sensitivity of RNS can be increased up to 80% by using different nerve-muscle combinations[12]. In a study comparing median and ulnar RNS, Pike-Lee et al. found that median decrement response (60.7%) was significantly higher than ulnar decrement response (35.7%), and concluded that it should be studied routinely[15]. In another study comparing the diagnostic sensitivity of RNS based on the type of muscle, it was determined that the deltoid was the most sensitive muscle, and the trapezius and nasalis muscles were comparably sensitive. Nevertheless, the fact that the deltoid muscle is more open to artifact and is painful to work with were cited among the disadvantages of working with deltoid muscle[4]. The importance of the clinically affected muscle has been emphasized in the context of RNS in the literature, and it was reported that a proximal muscle should be added to the study in the presence of ocular MG or bulbar involvement16. In comparison, the sensitivity (77.4%) of the trapezius muscle with spinal accessory nerve stimulation was found to be significantly higher than nasalis and ADM muscles. Therefore, although RNS is easily applicable from the ADM muscle and tolerated well by patients when used in the diagnosis of MG, it should not be forgotten that it has low diagnostic sensitivity. Hence, the trapezius muscle should be routinely used in the diagnosis of MG by RNS.

In parallel with the literature[17], anti-AChR and and anti-MuSK antibody test positivity were found in 63.6% and 6.3% of the patients, respectively. Conversely, no significant correlation was found between antibody positivity and the decremental response rate derived from all three muscles. In addition, there was also no significant relationship between the MGFA scores and decrement positivity. The number of patients with anti-MuSK positivity or seronegative patients in this study was quite limited. Accordingly, the study results mostly reflect the situation in AChR antibody-positive patients.

Sommer et al. performed trapezius muscle RNS test in patients with ocular-onset MG and found that the patients with positive decrement response progressed to generalized myasthenia at a significantly higher rate than those with negative decrement response. For this reason, they thought that RNS may also have a role in determining the prognosis[18]. In comparison, the analysis of the decrement responses of ocular-onset MG patients included in this study revealed that the decrement response from the trapezius was positive in 8 (61.5%) of the 13 patients. This finding suggests that the disease can become generalized regardless of whether it starts with ocular findings, thus that the

trapezius muscle RNS should definitely be studied to predict the transition.

In this study, we utilized a 10% decrement response as the threshold for RNS in diagnosing MG, a decision underpinned by a comprehensive review of existing literature and clinical practices. The 10% threshold is a well-established criterion in the diagnosis of MG, rooted in both historical and contemporary clinical practices. This benchmark is widely recognized due to its historical use and validation in numerous studies. For instance, Benatar and Kaminski highlight the traditional role of the 10% decrement in RNS as a diagnostic criterion for MG[19]. Additionally, the choice of the 10% threshold reflects a balance between sensitivity and specificity. While lower thresholds can increase sensitivity, they potentially compromise specificity. A study by Sanders et al. demonstrates the need to balance these two aspects in neuromuscular diagnostics[20]. It is important to note that the selection of any threshold value, including 10%, involves trade-offs. As evidenced by Abraham et al., exploring different threshold values like 6-7% can yield higher sensitivity, albeit with an increased risk of false positives[5]. In conclusion, our decision to utilize the 10% threshold for RNS in this study is grounded in a tradition of clinical and research practice, a balanced approach to sensitivity and specificity, and the need for standardized criteria in the field of neuromuscular disorders.

Major limitations of this research are its limited sample size and the lack of post-exercise facilitation in the methodology. Post-exercise facilitation refers to the improved response of muscles to nerve stimulation following a brief period of exercise. In MG, where nerve signal transmission to muscles is impaired, this phenomenon can be diagnostically significant. Post-exercise facilitation can enhance decremental response, making it more evident and aiding in diagnosis, especially in milder or early-stage cases[21].While valuable, our study did not focus on post-exercise facilitation but rather on assessing RNS sensitivity at rest across various muscles. This approach is supported by evidence suggesting a higher overall diagnostic efficacy when examining multiple muscles compared to relying solely on post-exercise facilitation[14,21]. Conversely, key strengths of this research lie in its forward-looking, prospective approach, selection of consecutive MG patients, and repetition of each measurement for 3 times.

CONCLUSION

In conclusion, the findings of this study revealed that although SF-EMG is the gold standard in the diagnosis of MG, RNS can also be used with high sensitivity provided that muscle selections are made accurately, particularly in cases where SF-EMG cannot be performed due to technical difficulties. Given its high sensitivity, the spinal accessory nerve should be included in routine RNS studies, particularly in the event of ocular-onset myasthenia. RNS can provide guidance in determining the prognosis in cases presenting with isolated ptosis or diplopia.

Ethical approval: Approval was obtained from local ethics committee (12.9.22/274). The procedures used in this study adhere to the tenets of the Declaration of Helsinki. Informed consent was obtained from all individual participants included in the study.

Conflict of Interest: The authors declared no conflicts of interest.

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REFERENCES

1. Lazaridis K, Tzartos SJ. Autoantibody Specificities in Myasthenia Gravis; Implications for Improved Diagnostics and Therapeutics. Front Immunol 2020;11:212. doi:10.3389/fimmu.2020.00212

2. Nic Hehir MK, Silvestri NJ. Generalized Myasthenia Gravis: Classification, Clinical Presentation, Natural History, and Epidemiology. Neurol Clin 2018;36(2):253-60. doi:10.1016/j.ncl.2018.01.002

3. AAEM Quality Assurance Committee. American Association of Electrodiagnostic Medicine. Literature review of the usefulness of repetitive nerve stimulation and single fiber EMG in the electrodiagnostic evaluation of patients with suspected myasthenia gravis or Lambert-Eaton myasthenic syndrome. Muscle Nerve 2001;24(9):1239-47. doi:10.1002/mus.1140

4. Amandusson Å, Elf K, Grindlund ME, Punga AR. Diagnostic Utility of Repetitive Nerve Stimulation in a Large Cohort of Patients With Myasthenia Gravis. J Clin Neurophysiol 2017;34(5):400-7. doi:10.1097/WNP.0000000000000398

5. Abraham A, Alabdali M, Alsulaiman A, et al. Repetitive nerve stimulation cutoff values for the diagnosis of myasthenia gravis. Muscle Nerve n2017;55(2):166-70. doi:10.1002/mus.25214

6. Stålberg E, van Dijk H, Falck B, et al. Standards for quantification of EMG and neurography. Clin Neurophysiol 2019;130(9):1688-729. doi:10.1016/j.clinph.2019.05.008

7. Sanders DB, Arimura K, Cui L, et al. Guidelines for single fiber EMG. Clin Neurophysiol 2019;130(8):1417-39. doi:10.1016/j.clinph.2019.04.005

8. Benatar M. A systematic review of diagnostic studies in myasthenia gravis. Neuromuscul Disord 2006;16(7):459-67. doi:10.1016/j.nmd.2006.05.006

9. Juel VC. Single fiber electromyography. Handb Clin Neurol 2019;160:303-10. doi:10.1016/B978-0-444-64032-1.00019-9

10. Giannoccaro MP, Paolucci M, Zenesini C, et al. Comparison of ice pack test and single-fiber EMG diagnostic accuracy in patients referred for myasthenic ptosis. Neurology 2020;95(13):e1800-e1806. doi:10.1212/WNL.0000000000010619

11. Mercelis R, Merckaert V. Diagnostic utility of stimulated single-fiber electromyography of the orbicularis oculi muscle in patients with suspected ocular myasthenia. Muscle Nerve 2011;43(2):168-70. doi:10.1002/mus.21853

12. Yildirim A, Adatepe T, Gunduz A, Yagiz O, Uzun N. Occipitalis muscle: using for repetitive facial nerve stimulation in myasthenia gravis. Acta Neurol Belg 2020;120(3):609-13. doi:10.1007/s13760-018-0889-9

13. Jaretzki A 3rd, Barohn RJ, Ernstoff RM, et al. Myasthenia gravis: recommendations for clinical research standards. Task Force of the Medical Scientific Advisory Board of the Myasthenia Gravis Foundation of America. Ann Thorac Surg 2000;70(1):327-34. doi:10.1016/s0003-4975(00)01595-2

14. Costa J, Evangelista T, Conceição I, de Carvalho M. Repetitive nerve stimulation in myasthenia gravis--relative sensitivity of different muscles. Clin Neurophysiol 2004;115(12):2776-82. doi:10.1016/j.clinph.2004.05.024

15. Pike-Lee T, Higginbotham D, Li Y. Direct comparison of median and ulnar repetitive nerve stimulation in generalized myasthenia gravis. Muscle Nerve 2021;64(4):490-3. doi:10.1002/mus.27366

16. Niks EH, Badrising UA, Verschuuren JJ, Van Dijk JG. Decremental response of the nasalis and hypothenar muscles in myasthenia gravis. Muscle Nerve 2003;28(2):236-8. doi:10.1002/mus.10411

17. 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. doi:10.1016/j.clinph.2011.02.026

18. Sommer N, Sigg B, Melms A, et al. Ocular myasthenia gravis: response to long-term immunosuppressive treatment. J Neurol Neurosurg Psychiatry 1997;62(2):156-62. doi:10.1136/jnnp.62.2.156

19. Benatar M, Kaminski HJ; Quality Standards Subcommittee of the American Academy of Neurology. Evidence report: the medical treatment of ocular myasthenia (an evidence-based review): report of the Quality Standards Subcommittee of the American Academy of Neurology. Neurology 2007;68(24):2144-149. doi:10.1212/01.wnl.0000263481.14289.90

20. Sanders DB, Wolfe GI, Narayanaswami P. Developing treatment guidelines for myasthenia gravis. Ann N Y Acad Sci 2018;1412(1):95-101. doi:10.1111/nyas.13537

21. Rubin DI, Hentschel K. Is exercise necessary with repetitive nerve stimulation in evaluating patients with suspected myasthenia gravis?. Muscle Nerve 2007;35(1):103-6. doi:10.1002/mus.20667