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EFFECTS OF ENVIRONMENTAL AND WEATHER CONDITIONS ON MYASTHENIA GRAVIS – IN SEARCH OF THE MISSING LINK

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Abstract

Ambient temperature can modulate neurotransmission in the peripheral nervous system. Transmission at the neuromuscular junction is also influenced by ambient temperature. Myasthenia Gravis (MG) is an autoimmune disease of the neuromuscular junction characterized by impaired transmission of the nerve impulses to the muscle fibers. The aim of the study was to determine if there is in fact a statistically significant correlation between environmental fluctuations and MG exacerbations among myasthenic patients. Also, the authors intended to find candidate variables for validation in a future prospective study. The subjects included in the study were hospitalized in I Neurology Clinic of "Prof. Dr. N. Oblu" Emergency Clinical Hospital Iasi, Romania during a five years period. The authors performed a binary logistic regression on 195 hospital admissions from which 144 were from Iasi County. P values<0.05 were considered significant. The risk of myasthenic aggravation appeared to be higher when the extremes of temperature from the week before hospitalization were greater with a $p < 0.001$ and Exp(B) of 1.143 (95%CI: 1.1 - 1.187). This statistically significant correlation maintained when a subgroup analysis of 144 admissions from Iasi county was performed. Also, apparently females had a greater risk of exacerbation when ambient temperature fluctuations were higher. These are promising remarks that represent a hypothesis for a prospective study. In addition, further studies are mandatory in order to establish a link between myasthenic aggravations and other environmental parameters such as atmospheric pressure and humidity.

Keywords: aggravation, extremes of temperature, myasthenia gravis, temperature range

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1. Introduction

The environment plays an important role in the normal functioning of the human organism. Both chemical and physical factors influence human organs and systems at their most basic level such as homeostatic equilibrium and neurotransmission. Any imbalance in these factors causes or contributes to systemic diseases. A classic example is soil and water iodine deficiency enhanced by deforestation and the appearance of thyroid enlargement and other iodine deficiency disorders (Preda et al., 2013). Other examples address allergic asthma secondary to exposure to environmental vegetal dust which can appear as an occupational or related profession disease (Constantin et al., 2015), health risks for patients with hepatitis A virus (Vata et al., 2018), environment for *Mycobacterium tuberculosis* growth (Cislariu et al., 2018), particulate matter air pollution effects on vulnerable and non-vulnerable people (Noor et al., 2015; Oprea et al., 2017).

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Neurotransmission is the basis of communication between neurons and their targets. Information to other neurons or effector cells is transferred through electrical and chemical synapsis, the last being the most frequent. Influence of environmental factors, especially ambient temperature on neurotransmission velocity in the peripheral nervous system has been a subject of interest since the experimental studies published in the early 1900 (Gasser, 1928). High temperature non-linearly increases conduction velocity in motor and sensory fibres (Rutkove et al., 1997), while cold environments have an opposite effect (Todnem et al., 1989).

The complex chemical synapse between peripheral nerve endings and striated muscle fibres can be the target of several neuromuscular junction diseases, the most studied being myasthenia gravis (MG) (Vincent et al., 2001) Also known as Erb Goldflam disease, non-hereditary MG has a worldwide incidence which varies from 0.25 to 2 per million persons (Carr et al., 2010). In the last decades its prevalence has increased due to both prolonged life expectancy and also advances in diagnosis methods (Melzer et al., 2016). MG is an autoimmune disorder in which auto-antibodies attack the motor end-plate resulting in an impaired transmission of the nerve impulses to the muscle fibers (Ramanujam et al., 2011).

In 85% of cases, the target of the auto-antibodies is the acetylcholine receptor (Lindstrom, 2000) followed by the muscle-specific tyrosine kinasereceptor (Hoch et al., 2001). In 7% of seronegative myasthenic patients another antibody is detected, against lipoprotein related protein 4 (Romi et al., 2017). MG associated with timus neoplasia occurs in 10-15% of myasthenic cases and it is asociated with other auto-antibodies (Evoli et al., 2002).

MG has multiple clinical and paraclinical subtypes that may differ regarding prognosis and therapeutical approach. MG has an ocular and a general form. The ocular form is more frequent among people of asian descend (Zhang et al., 2007). The general form involves all striated muscles. It can appear before or after the age of 50 (Somnier, 2005; Suzuki et al., 2011). From this point of view, MG can be early-onset (EOMG) and late-onset (LOMG). For therapeuticall and general outcome reasons the Task Force of the Medical Advisory Board of the Myasthenia Gravis Foundation Of America (MGFA) created a five grade clinical scale using Osserman's original stadialization as a model (Jaretzki et al., 2000).

According to this classification MG can be divided into five main classes and several subclasses: class I defines the ocular form whereas classes II to V, generalized forms in various degrees of severity. Each of the latter is subdivided in type A defined as predominantly limb and/or axial muscles and type B defined as predominantly bulbar and/or respiratory muscles.

Usualy MG has a prolonged and fluctuating evolution: in the first years relapses alternate with remission periods while in much more advance stages, relapses tend to be more scarce and a moderate motor deficit may be present permanently. Relapses can be spontaneus or can be triggered by pregnancy, sistemic infections or certain medication which alteres the normal transmission at the neuromuscular junction (Oosterhuis, 1989).

The clinical aspect of a relapse can vary from exacerbation of double vision or of generalized fatigability to life-threatening myasthenic crisis. Differential diagnosis varies with the clinical pattern of an exacerbation and can include ischemic or hemorrhagic stroke, central nervous system neoplasia, myopathies and encephalitis. One of the most controversial pathologies from the latter category is Herpes Simplex encephalitis which can mimic a myasthenic aggravation. According to Boangher, particular forms of Herpes simplex encephalitis with fluctuating neurological deficits might be a consequence of a secondary autoimmune phenomenon related to the presence of NMDA-R antibodies (Boangher et al., 2018). Myasthenic crisis is characterized by acute respiratory insufficiency and hemodynamic instability. Therefore, it can be easily mistaken with other causes of acute respiratory insufficiency. Among these, even though rare, one must not ignore the possibility of an autonomic malfunction secondary to a lesion in the vegetative nervous system, either central or peripheric. In right handed patients, for example, it has been shown that a middle cerebral artery ischemic stroke is associated with different hart rate responses depending on the side of the lesion: right hemisphere infarcts have an enhanced sympathetic control on the heart rate while left hemispheric ones have a dominant parasympathetic control (Constantinescu et al., 2016, 2018).

In vivo, peripheral nerves form a functional unit with striated muscle fibres using the neuromuscular junction as an interface between the presynaptic and postsynaptic component. At the synaptic site the endplate potential is converted into muscle action potential (Verschuuren et al., 2016). Therefore, the effects of ambient temperature on the peripheric nerves are evident also in the muscle function.

Several electrophysiological studies have demonstrated influence of temperature on neuromuscular transmission in MG. Overall, local cooling improves muscular transmission (Borenstein and Desmedt, 1975) whereas local heating reduces the amplitude of the action potential (Racinais and Oksa, 2010). These facts support the much older clinical observation that myasthenic symptoms, especially muscle weakness and palpebral ptosis may improve with cold and may worsen with heat (Simpson, 1960). Both clinical observation and electrophysiological proof suggest that ambient temperature may influence the symptomatology of MG and even weather can have clinical implications (Borenstein and Desmedt, 1974). There is cited an increase in acute exacerbation in late summer and late winter (Melamed et al., 2014).

In spite of these correlations few data are available regarding seasonal patterns of MG worldwide, even fewer are reported from a specific region in Romania. Overall there are also few data regarding occurrence of MG in Eastern European countries (Ziedaa et al., 2018). Iasi is major city in north-eastern Romania and the existence of university neurology clinics explains a high addressability of the myasthenic patients. Also, the city possess an important meteorological center – The Moldova Regional Meteorological Centre.

The purpose of this study was to determine if there is in fact a correlation between environmental temperature fluctuations and MG exacerbations among myasthenic patients which were hospitalized in I Neurology Clinic of "Prof. Dr. N. Oblu" Emergency Clinical Hospital Iasi. In addition, finding candidate variables for validation in a future prospective study was also a priority.

2. Material and methods

2.1. Data acquisition

Patient data were collected from patient files of I Neurology Clinic from "Prof. Dr. N. Oblu" Emergency Clinical Hospital Iasi, hospitalized between 1.01.2013 and 31.12 2017. Both prevalent cases and incident cases with a positive diagnosis of MG were included in the study. As stipulated in the medical literature, a positive case was defined by suggestive clinical aspect along with elevated specific autoantibodies titers and/or positive repetitive nerve stimulation test. The patients which had no significant decrement in spite of characteristic symptoms underwent a single fiber electromyography test or jitter test.

According to standardized methodology published in medical literature, low frequency repetitive nerve stimulation test consists of applying electrical current on a motor nerve with a frequency of two to five Hertz. Five cycles of ten stimuli each are applied. At this low frequency, presynaptic depression is greater than acetylcholine release facilitation. After stimulation the difference between negative waves' amplitude of the first and the forth stimuli is calculated. A decrement at least equal to 10% is considered highly suggestive for MG.

Environmental data were obtained from The Moldova Regional Meteorological Centre, Iasi and included daily values of maximum and minimum temperatures from 1.01.2013 and 31.12 2017 for Iasi city. A time interval of two weeks before an aggravation was taken into consideration. Six parameters were calculated: standard deviation for temperatures in the week prior to hospitalization (DST1) and the week before that (DST2), weekly temperature range defined as the difference between the highest and the lowest temperature from the week prior to hospitalization (RT1) and the week before that (RT2) and the average of the temperatures within the week prior to hospitalization (MT1) and the week before that (MT2).

2.2. Statistical analysis

One sided Fisher's exact test was used for comparison of categorical data. Mantel-Haenszel test was used for comparison of stratified categorical data. In order to verify the potential influence of the six weather parameters taken into consideration on myasthenic aggravations we used conditional forward binary logistic regression without intercept.

The reason of admission was considered the dependent variable and was given 1 for aggravation and 0 for assessment (stable disease). None of the independent variables were separately analyzed before they were introduced in the model, nor they were divided by category. P values<0.05 were considered significant. Excel version 1811 was used for data management and statistical analysis was performed using SPSS 20.

3. Results and discussions

3.1. Clinical data

In the period included in the study, a total of 122 myasthenic patients (83 females and 39 males) were registered from which 79 were incident cases: 22 in 2013, 17 in 2014, 14 in 2015, 14 in 2016 and 12 in 2017. The patients lived in Iasi city, Iasi county or neighbor counties. The median age at first presentation was 51 (range 19-83; mean 49) for women and 62 (range 31-78; mean 61) for men.

Regarding age at onset and gender, 51 of the cases had an age at onset under 50 years, from which 45 were women. From the 71 patients that were over 50 years old at the time of onset, 33 were men and 38 were women (Fig. 1). The clinical characteristics of the subjects are presented in Table 1 using MGFA modified Osserman classification.

During the specific time interval taken into consideration 105 patients were admitted for worsening of symptoms. Overall, a total of 195 admissions with 155 aggravations were noted (Table 2). From these, 144 admissions were from Iasi County. Figs. 2 and 3 reveal gender and age distribution among the 195 admissions. Female aggravations represented 83.58% of total female admissions while male aggravations represented 70.49% of total male hospitalizations. When age was used as stratifying factor for gender and admission reason distributions no statistically significant difference was found when using Mantel-Haenszel Mantel-Haenszel test.

There is no unanimous opinion regarding a certain age limit wich divides MG in early onset and late onset: some authors consider the age of 40, whereas others raise the limit to 60 years. However, the current tendency is to establish the age of 50 as the treshold (Somnier, 2005). EOMG is tipically associated with females, timus hiperplasia and high

level of acetylcholine receptor antibodies, whereas LOMG tends to be more frequent in men which do not have any timus modification (Berrih-Aknin et al., 2014; Meriggioli and Sanders, 2009). The lot taken into consideration, representing both prevalent and incident cases of MG admitted in a university hospital in a major north-eastern city of Romania during a five year period tends to fit in the already well-established worldwide epidemiology.

More specifically, the EOMG cases were predominantly women. Even though in the LOMG subgroup women were also the majority, the fact that the total number of men was 39, from witch 33 had over 50 years at the time of MG onset suggests that men are more prone to develop the disease later in life.

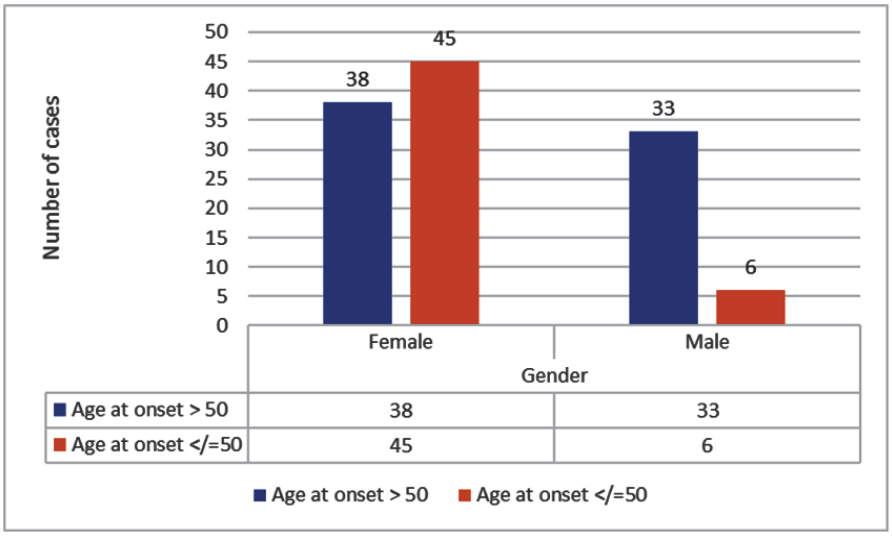

Fig. 1. 122 MG patients: gender and age at onset ($p < 0.001$, one sided Fisher's exact test)

Table 1. MGFA modified Osserman classification

Osserman	*Female*		*Male*		*Total*
	Count	*% of total*	*Count*	*% of total*	
Osserman I	22	75.9	7	24.1	29
Osserman IIA	30	69.8	13	30.2	43
Osserman IIB	26	61.9	16	38.1	42
Osserman IIIA	2	100	0	0	2
Osserman IIIB	1		2		3
Osserman IVA	1	50	1	50	2
Osserman IVB	1	100	0	0	1
Osserman V	0	0	0	0	0

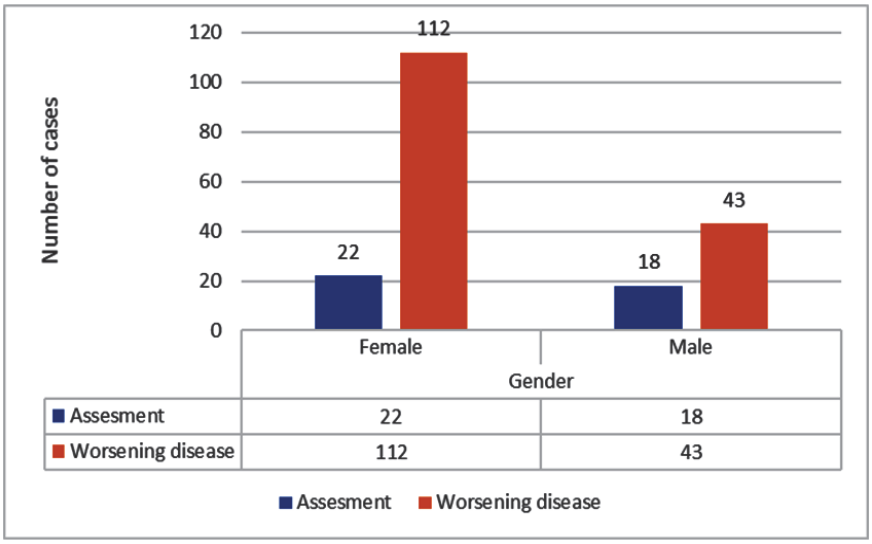

Fig. 2. Distribution of events according to gender and admission reason
(One sided Fisher's exact test $p = 0.03$)

Table 2. Breakdown of admissions

			Year					*Total*
			2013	***2014***	***2015***	***2016***	***2017***	
Worsening disease	Month	January	4	1	2	3	5	15
		February	3	2	3	6	7	21
		March	4	4	1	3	3	15
		April	7	6	5	3	2	23
		May	2	1	0	4	3	10
		June	3	1	1	1	2	8
		July	2	0	3	1	2	8
		August	3	1	3	2	3	12
		September	5	4	2	1	2	14
		October	3	3	2	3	2	13
		November	2	2	1	1	2	8
		December	1	6	0	1	0	8
	Total		**39**	**31**	**23**	**29**	**33**	**155**
			Year					*Total*
			2013	***2013***	***2013***	***2013***	***2013***	
Assessment	Month	January	0	0	0	0	0	0
		February	0	0	0	1	1	2
		March	0	0	1	3	3	7
		April	0	0	0	4	2	6
		May	0	1	0	1	2	4
		June	1	2	0	0	1	4
		July	0	0	0	2	3	5
		August	0	1	0	0	1	2
		September	0	1	1	0	0	2
		October	1	0	1	0	0	2
		November	0	1	1	0	3	5
		December	0	0	0	1	0	1
	Total		**2**	**6**	**4**	**12**	**16**	**40**

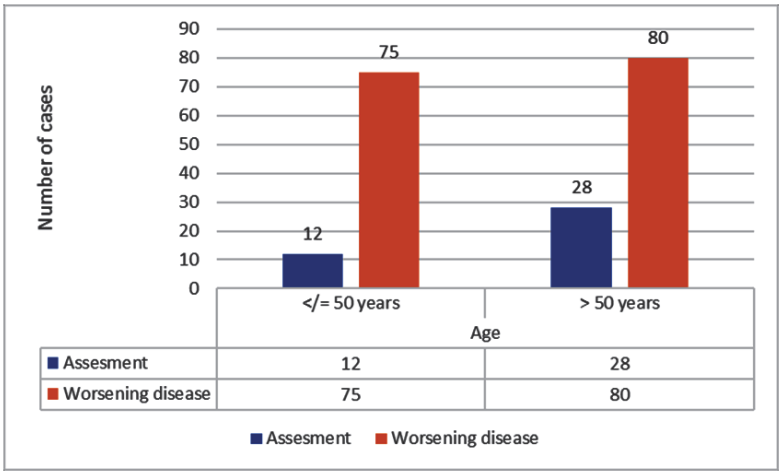

Fig. 3. Distribution of events according to age and admission reason
(One sided Fisher's exact test p=0.027)

3.2. Meteorological implications

3.2.1. Entire lot

Few data are available regarding duration of exposure to a certain environmental temperature or pressure and myasthenic symptoms fluctuations in MG. The time interval of two weeks before an aggravation was taken into consideration in order to cover possible influences of temperature fluctuations on myasthenic aggravations. During the time period from 1 January 2013 to 31 December 2017, from all six meteorological parameters taken into consideration, RT from the last week prior to month of hospitalization has correlated statistically significant with exacerbation of symptoms with a $p < 0.001$ and Exp(B) of 1.143 (95%CI: 1.1 - 1.187). Tables 3 and 4 reveal all six independent variables and their level of significance. When gender variable was also taken into account, both gender and DST1 correlated significantly with exacerbation of symptoms (Table 5).

3.2.2. Subgroup analysis

Given the fact that from the entire lot of 195 admissions 144 were from Iasi County, a subgroup analysis was performed. When only 144 admissions from Iasi county were taken into account and all six temperature variables were used (without gender), again RT1 has correlated statistically significant with exacerbation of symptoms with $p < 0.001$ and exp(B) of 1.112 (95%CI: 1.068 – 1.158) (Table 6). After adding gender, female gender was the only variable to corelate with exacerbation of symptoms with $p<0.001$ and exp(B) of 3.947 (95%CI: 2.386-6.53) (Table 7).

Remarks towards the statistical analysis performed are mandatory. Even though the type of analysis performed has an increased overfitting risk, the authors consider the results useful for generating hypothesis to be tested in a prospective study.

Including as much data as possible in order to obtain promising working hypothesis was the reason why six independent variables were introduced in the model. The present paper is one of the few that focuses on possible correlations between MG exacerbations and weather variables. In fact, most of the articles concerning this subject find lack correlation between weather, temperature and patient general muscle weakness. One of the explanations is the existence of other much more evident trigger factors like therapy, rest and exercise, administration of certain medication, infections. However, one of the first articles that pioneered the idea of weather influenced myasthenic symptoms reveals that extremes of heat and cold do make the fluctuation of symptoms more evident to the patient because he is stressed by thermoregulation demands (Borenstein and Desmedt, 1974).

Table 3. Influence of RT1 on symptom exacerbations

		B	*S.E.*	*Wald*	*df*	*Sig.*	*Exp(B)*	*95% C.I.for EXP(B)*	
								Lower	*Upper*
Step 1	RRT1	.133	.019	47.763	1	.000	1.143	1.100	1.187

-2 Log Likelihood: 207.481; Nagelkerke R^2: 0.367

Table 4. Variables not introduced in the model

			Score	*df*	*Sig.*
Step 1	Variables	DST1	.747	1	.387
		MT1	.449	1	.503
		DST2	.707	1	.400
		RT2	1.163	1	.281
		MT2	.576	1	.448
	Overall Statistics		2.910	5	.714

Table 5. Influence of gender and temperature variability on symptom exacerbations

		B	*S.E.*	*Wald*	*df*	*Sig.*	*Exp(B)*	*95% C.I.for EXP(B)*	
								Lower	*Upper*
Step 1[a]	Sex(1)	1.627	.233	48.703	1	.000	5.091	3.223	8.041
Step 2[b]	DST1	.195	.072	7.226	1	.007	1.215	1.054	1.401
	Sex(1)	1.046	.310	11.379	1	.001	2.846	1.550	5.225
a. Variable(s) entered on step 1: Sex.									
b. Variable(s) entered on step 2: DST1b.									

-2 Log Likelihood: 196.304; Nagelkerke R^2: 0.421

Table 6. Events from Iasi County - influence of temperature variables

		B	*S.E.*	*Wald*	*df*	*Sig.*	*Exp(B)*	*95% C.I.for EXP(B)*	
								Lower	*Upper*
Step 1[a]	RRT1	.107	.021	26.859	1	.000	1.112	1.068	1.158

a. Variable(s) entered on step 1: RT1c.

-2 Log Likelihood: 167.353; Nagelkerke R^2: 0.268

Table 7. Events from Iasi County. Influence of temperature variables and gender

		B	*S.E.*	*Wald*	*df*	*Sig.*	*Exp(B)*	*95% C.I. for EXP(B)*	
								Lower	*Upper*
Step 1[a]	Sex(1)	1.373	.257	28.580	1	.000	3.947	2.386	6.530
a. Variable(s) entered on step 1: Sex.									

-2 Log Likelihood: 163.942; Nagelkerke R^2: 0.293

Apparently, aggravations were influenced only by RT and DST from the last week prior to month of hospitalization. The results obtained are consistent with the ones from the medical literature. In both the lot from Iasi County and also the entire lot, myasthenic exacerbations tended to be more frequent as the difference between the highest and the lowest temperature from that week was higher. In other words, as the extremes of temperature are greater, the thermoregulation effort is increased and this has a negative impact on the transmission at an already impaired neuromuscular junction. The result is a higher number of exacerbations.

Regarding gender related exacerbations, in the entire lot females tended to have an increased probability of aggravation when ambient temperature fluctuations were higher. This could have been a bias do to the fact that women represented the majority of the lot. However, if female aggravations related to total female admission are compared to men aggravation related to total male admissions, a tendency for female aggravations predominance is still obvious.

When the same reasoning was applied in the Iasi county subgroup, again females appeared to have a much higher rate of exacerbation. DST 1 and RT1 variables were at the limit of statistical significance. Even though this suggested that the higher the ambient temperature fluctuations were, the greater the risk of exacerbation in women was, unfortunately no such conclusion can be drawn due to the fact p value is above the threshold considered.

4. Conclusions

The present paper consists in a valid appraisal of possible influence of temperature fluctuations on MG aggravations.

In the lot considered the risk of myasthenic aggravation appears to be higher when the extremes of temperature from the week before hospitalization were greater. This finding has a potential great impact in the general management of MG. To be more specific, along the general dietary and hygienic recommendations that neurologist give to their myasthenic patients, advices to avoid exposure to very low or very high environmental temperatures can also be formulated. These behavioural precautions may be highlighted especially in women suffering from MG. This is because, according to the current study, apparently females have a greater risk of exacerbation when ambient temperature fluctuations were higher.

These are promising remarks that represent a hypothesis for a prospective study. If a certain cause-effect relation is demonstrated between environmental temperature fluctuations and myasthenic aggravations in a much larger lot the implications would be extraordinary in terms of both prophylaxis and treatment of MG. In other words, myasthenic aggravations could be attenuated or ideally avoided if the patient would not expose himself to extreme environmental temperatures. Also, if an exposure to a low or elevated temperature is anticipated then a myasthenic exacerbation could be avoided by temporarily modifying the medication posology.

In addition, further studies are mandatory in order to establish a link between myasthenic aggravations and other environmental parameters such as atmospheric pressure and humidity. The medical implications are extensive: myasthenic patients could prevent an exacerbation by avoiding exposure to certain weather conditions which could have a positive impact on reducing medication and improving quality of life and overall outcome.

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References

Berrih-Aknin S., Le Panse R., (2014), Myasthenia gravis: a comprehensive review of immune dysregulation and etiological mechanisms, *Journal of Autoimmunity*, **52**, 90-100.

Boangher S., Mespouille P., Sophie G., Van Pesch V., Cuciureanu D., (2018), Herpes simplex encephalitis relapse associated with positive N-methyl-D-aspartate receptor antibodies, *Acta Neurologica Belgica*, **118**, 1-3.

Borenstein S., Desmedt J.C., (1974), Temperature and weather correlates of myasthenic fatigue, *Lancet*, **304**, 63-66.

Borenstein S., Desmedt J.C., (1975), Local cooling in myasthenia. Improvement of neuromuscular failure, *Archives of Neurology*, **32**, 152-157.

Carr A.S., Cardwell C.R., McCarron P.O., McConville J.A., (2010), Systematic review of population based epidemiological studies in myasthenia gravis, *BMC Neurology*, **46**, 1471-2377.

Cislariu S.A., Lacatusu G.A., Largu A., Iordan I.F., Vata A., Manciuc C., (2018), Changes in glucose levels – a predictive marker for an adequate environment aimed at *Mycobacterium tuberculosis* growth, *Environmental Engineering and Management Journal*, **17**, 3007-3011.

Constantin B., Postolache P., Croitoru A., Nemes R., (2015), Occupational bronchial asthma - Clinical and epidemiological aspects, *Journal of Environmental Protection and Ecology*, **16**, 517-520.

Constantinescu V., Matei D., Costache V., Cuciureanu D., Arsenescu-Georgescu C., (2018), Linear and nonlinear parameters of heart rate variability in ischemic stroke patients, *Neurologia I Neurochirurgia Polska*, **52**, 194-206.

Constantinescu V., Matei D., Cuciureanu D., Corciova C., Ignat B., Popescu C.D., (2016), Cortical modulation of cardiac autonomic activity in ischemic stroke patients, *Acta Neurologica Belgica*, **116**, 473-480.

Evoli A., Minisci C., Di Schino C., Marsili F., Punzi C., Batocchi A.P., Tonali P.A., Doglietto G.B., Granone P., Trodella L., Cassano A., Lauriola L., (2002), Thymoma in patients with MG: characteristics and long-terme outcome, *Neurology*, **59**, 1844-1850.

Gasser H.S., (1928), The relation of the shape of the action potential of nerve to condition velocity, *American Journal Physiology*, **84**, 699-711.

Hoch W., McConville J., Helms S., Newsom-Davis J., Melms A., Vincent A, (2001), Auto-antibodies to the receptor tyrosine kinase MuSK in patients with myasthenia gravis without acetylcholine receptor antibodies, *Nature Medicine*, **7**, 365-368.

Jaretzki A., Barohn R.J., Ernstoff R.M., Kaminski H.J., Keesey J.C., Penn A.S., Sanders D.B., (2000), Myasthenia gravis: recommendations for clinical research standards. Task force of the medical scientific advisory board of the myasthenia Gravis Foundation of America, *Neurology*, **55**, 16-23.

Lindstrom J.M., (2000), Acetylcholine receptors and myasthenia, *Muscle & Nerv*, **23**, 453-477.

Melamed R.D., Khiabanian H., Rabadan R., (2014), Data-driven discovery of seasonally linked diseases from an Electronic Health Records system, *BMC Bioinformatics*, **15**, S3, https://doi.org/10.1186/1471-2105-15-S6-S3.

Melzer N., Ruck T., Fuhr P., Gold R., Hohlfeld R., Marx A., Melms A., Tackenberg B., Schalke B., Schneider-Gold C., Zimprich F., Meuth S.G., Wiendl H., (2016), Clinical features, pathogenesis, and treatment of myasthenia gravis: a supplement to the Guidelines of the German Neurological Society, *Journal of Neurology*, **263**, 1473-1494.

Meriggioli M.N., Sanders D.B., (2009), Autoimmune myasthenia gravis: emerging clinical and biological heterogeneity, *The Lancet Neurology*, **8**, 475-90.

Noor N.M., Yahaya A.S., Ramli N.A., Luca F.A., Al Bakri Abdullah M.M., Sandu A.V., (2015), Variation of air pollutant (particulate matter - PM10) in Peninsular Malaysia Study in the southwest coast of peninsular Malaysia, *Revista de Chimie*, **66**, 1443-1447.

Oosterhuis H.J., (1989), The natural course of myasthenia gravis: a long term follow up study, *Journal Neurology Neurosurgery Psychiatry*, **52**, 1121-1127.

Oprea M., Dunea D., Liu H.-Y., (2017), Development of a knowledge based system for analyzing particulate matter air pollution effects on human health, *Environmental Engineering and Management Journal*, **16**, 669-676.

Preda C., Ungureanu M., Leuştean L., Cristea C., Mogos V., Vulpoi C., Gavrilescu M., (2013), Human health related to iodine environmental occurrence and its deficiency in water and food, *Environmental Engineering and Management Journal*, **12**, 1045-1049.

Racinais S., Oksa J., (2010), Temperature and neuromuscular function, *Scandinavian Journal of Medicine Science Sports*, **20**, 1-18.

Ramanujam R., Piehl F., Pirskanen R., Gregersen P.K., Hammarstrom L., (2011), Concomitant autoimmunity in myasthenia gravis – Lack of association with IgA deficiency, *Journal of Neuroimmunology*, **236**, 118-122.

Romi F., Hong Y., Gilhus N.E., (2017), Pathophysiology and immunological profile of myasthenia gravis and its subgroups, *Current Opinion in Immunology*, **49**, 9-13.

Rutkove S.B., Kothari M.J., Shefner J.M., (1997), Nerve, muscle, and neuromuscular junction electrophysiology at high temperature, *Muscle Nerve*, **20**, 431-436.

Simpson J.A., (1960), Myasthenia gravis: a new hypothesis, *Scottish Medical Journal*, **5**, 419-436.

Somnier F.E., (2005), Increasing incidence of late-onset anti-AchR antibody-seropositive myasthenia gravis, *Neurology*, **65**, 928-930.

Suzuki S., Utsugisawa K., Nagane Y., Satoh T., Kuwana M., Suzuki N., (2011), Clinical and immunological differences between early-onset and late-onset myasthenia gravis in Japan, *Journal of Neuroimmunology*, **230**, 148-152.

Todnem K., Knudsen G., Riise T., Nyland H., Aarli J.A., (1989), The non-linear relationship between nerve conduction velocity and skin temperature, *Journal Neurology Neurosurgery Psychiatry*, **52**, 497-501.

Vâță A., Manciuc C., Dorobăț C., Vâță L.G., Luca C.M., (2018), biochemical investigations in the assessment of health risks for over 35-year-old patients affected by environments with hepatitis A virus, *Environmental Engineering and Management Journal*, **17**, 2749-2754.

Verschuuren J., Strijbos E., Vincent A., (2016*), Neuromuscular Junction Disorders*, In: *Handbook of Clinical Neurology*, Pittock S.J, Vincent A. (Eds.), Elsevier, New York, 447-466.

Vincent A., Palace J., Hilton-Jones D., (2001), Myasthenia Gravis, *Lancet*, **357**, 2122-2128.

Zhang X., Yang M., Xu J., Zhang M., Lang B., Wang W., Vincent A., (2007), Clinical and serological study of myasthenia gravis in HuBei Province, China, *Journal Neurology Neurosurgery Psychiatry*, **78**, 386-390.

Zieda A., Ravina K., Glazere I., Pelcere L., Naudina M.S., Liepina L., Kamsa I., Kurjane N., Woodhall M., Jacobson L., Leite M.I., Tandon K., Kenina V., (2018), A nationwide epidemiological study of myasthenia gravis in Latvia, *European Journal of Neurology*, **25**, 519-526.