deficiency exacerbates the atrophy induced by denervation, indicating a direct involvement of MSY3 in the muscle-nerve interaction. Since MSY3 controls proper myogenin expression along the muscle fiber and consequently the correct AChR distribution at the end-plates, our working hypothesis is that MSY3-myogenin-AChR and other myogenin targets is a crucial network that regulates NMJ structure and function, and muscle mass, in normal and diseased muscles. By analyzing the distribution of myofibers with an oxidative or glycolitic metabolism in muscles of old WT and hom MSY3 KO mice, we observed a strong increase of oxidative type fibers in the mutant hom mice, suggesting a role of this protein in muscle metabolism regulation, especially in elderly mice. Moreover, analysis of the MSY3 binding sites in adult muscle, shows a preferential occupancy in proximity of genes that control mitochondrial oxidative phosphorylation. We also have evidence that HDACs control MSY3 activity in response to nerve signals. Indeed upon denervation we observed a strong enrichment of the MSY3 occupancy at the myogenin promoter when HDAC4 is genetically ablated, suggesting a role of HDAC4 in inhibiting MSY3 DNA binding capacity. All together these results suggest that MSY3 can mediate modulation of the myogenin regulatory pathway during muscle atrophy and it is a potential therapeutic target for therapies for muscle degeneration-associated diseases.

MSY3/Csda, myogenin, neuromuscular junction, muscle wasting, denervation

Muscle function- #3015
P18- 282- Mechanosensing and mechanosignaling by caveolae: A new role in human muscular dystrophy diseases
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Cells perceive their microenvironment not only through signaling receptors, but also through physical and mechanical cues. Cells translate these stimuli by mechanotransduction into biochemical signals controlling multiple cellular functions. We recently established caveolae as mechano-sensors that play a major role in the homeostasis of the membrane tension of the cell membrane. Caveolae are 60-80 nm cup-shaped membrane invaginations, rich in sphingolipids and cholesterol and made of oligomerized caveolins (Cav1, 2 or 3). Caveolae are particularly abundant in muscle cells. We have investigated the role that caveolae play in the physical and biological responses to mechanical stress at the plasma membrane of human muscle cells. Based on membrane tension measurements and mechano-signaling responses to cyclic stretching, we show that caveolae are not mechanically functional in several limb-girdle muscular dystrophies. These studies open the way to a new mechanistic understanding of human muscular dystrophies and to the elaboration of new therapeutics to correct caveolae dysfunction in muscular dystrophies.

caveolae, mechanics, myotube, mechanosignaling

Muscle function- #4467
P18- 283- Muscle contraction is required to maintain the pool of muscle progenitors via the transcriptional co-activator YAP and NOTCH during fetal myogenesis
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Skeletal muscle is a plastic tissue that can adapt its size according to changes in mechanical loading. The variations in muscle mass can occur by changes in fibre size and/or by changes in satellite cell number (stem cells). The development, homeostasis and regeneration of skeletal muscle rely on progenitor/stem cells. Although, the respective contributions of muscle fibres and stem cells in muscle wasting in unloading conditions and disease-mediated atrophy is not well established.
We used the chick as a model to study the role of mechanical forces in the maintenance of skeletal muscle mass and progenitor cells during foetal myogenesis.
We demonstrated that the activity of NOTCH signalling pathway, known to be a central regulator of muscle stem cells, was decreased in foetal muscles following immobilization. Moreover, the inhibition of muscle contraction mimicked a NOTCH loss-of-function phenotype, i.e. dramatically decreased the number of foetal muscle progenitors (PAX7+ cells) that shifted towards a differentiation fate (increased MYOD and MYOG expression). Forced-NOTCH activation prevented the diminution in the number of muscle progenitors in immobilized embryos. We also provide evidence that immobilization reduces the amount of the transcriptional co-activator YAP in nuclei of post-mitotic muscle fibres and results in reduced JAG2 expression. Our results identify a novel mechanism acting downstream of mechanical forces and indicate that muscle activity signals via YAP and NOTCH to regulate the pool of foetal muscle progenitors.

P19 – Myasthenia (immune & congenital)- N° 284 to N° 298

Congenital myasthenic syndromes- # 2458
P19- 284- Identification of MYO9A as a novel causative gene in congenital myasthenic syndrome
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Background: Congenital myasthenic syndromes (CMS) are a group of genetically heterogeneous disorders characterised by compromised function at the NMJ. CMS manifests in childhood with fatigable weakness of limb, ocular and bulbar muscles.
Aims: To identify novel CMS genes by whole exome sequencing (WES).
Methods: DNA from a cohort of patients with a clinical diagnosis of CMS with suspected autosomal recessive inheritance was sent to deCODE genetics for WES. Variants in the exome were filtered to exclude those with a frequency greater than 1% in control populations, unlikely to significantly impact the protein structure and not compatible with the inheritance model. Genes that had not been excluded were then segregated within the families. Identified candidates were subject to functional analysis in

vitro and in vivo, utilising both shRNA mediated knockdown in cells and antisense morpholino oligonucleotide knockdown in zebrafish.

Results: MYO9A was identified as a potentially causative gene in two unrelated families. Immunofluorescent staining revealed co-localisation of MYO9A at the NMJ and knockdown of MYO9A in NSC-34 cells lead to a disruption in branching of sprouting neurites. Two orthologues for MYO9A were identified and targeted in zebrafish, causing abnormal movement in response to tactile stimulation and abnormal branching and shortening of motor axons.

Conclusion: MYO9A has been identified as a possible causative gene in CMS. The preliminary studies carried out in cell culture and zebrafish knockdowns demonstrated phenotypes consistent with an effect on the neuromuscular junction. To confirm results obtained using MO injection, we are currently utilising CRISPR technology to target the genes in zebrafish.

NMJ, unconventional myosin, CMS, zebrafish, Whole exome sequencing

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Congenital myasthenic syndromes- #2544

P19- 285- Mutations in GFPT1 and DPAGT1 identified in a French Cohort of Patient with congenital myasthenia cause glycosylation defects and neuromuscular junction abnormalities on muscle biopsies

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Limb-girdle congenital myasthenic syndrome (LG-CMS) forms a distinct group of CMS mainly characterized by prominent muscle weakness in proximal skeletal associated with sarcoplasmic reticulum reorganization (the so called tubular aggregates) visible in patient muscle biopsies. Mutations in GFPT1 (glutamine-fructose-6-phosphate transaminase 1) and DPAGT1 (dolichyl-phosphate N-acetyglucosamine transferase 1), two key enzymes involved in the fine tuning of glycosylation, are the main cause of this particular syndrome (Senderek et al., 2011; Belaya et al., 2012). However the consequences of a deficit in the ubiquitous N-glycosylation process of proteins at the neuromuscular junction (NMJ) remain largely unknown. In this study, we report on recessively inherited mutations in GFPT1 and DPAGT1 in a French cohort affected with LG-CMS with tubular aggregates. We collected clinical data of 6 patients from 5 families and identified 5 new and undescribed mutations in GFPT1 and 2 new undescribed mutations in DPAGT1. We carefully analyzed the NMJ in all patients both by whole-mount immunochemistry and at the ultrastructural level. We demonstrated that defects in GFPT1 and DPAGT1 induced a profound remodeling of endplates characterized by a progressive loss of junctional folds, abnormal nerve endings, as well as impaired glycosylation process associated with increased Peanut agglutinin (PNA) lectin suggesting a deficit in muscle protein glycosylation. This study shed light onto the etiopathology of this rare neuromuscular disorder which may be considered as a ?Tubular aggregate myopathy with synaptopathy?.

Congenital myasthenic syndromes, GFPT1, DPAGT1, neuromuscular junction, Tubular aggregates

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Congenital myasthenic syndromes- #2564

P19- 286- A GFPT1 deficient mouse model of Congenital Myasthenic Syndrome

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Congenital myasthenic syndromes (CMS) occur as a result of genetically inherited mutations giving rise to impaired transmission at the neuromuscular junction (NMJ). Patients with mutations in GFPT1 demonstrate a limb-girdle pattern of weakness, tubular aggregates in muscle biopsies, and an unusual sparing of the ocular, facial and bulbar muscles. GFPT1 encodes a ubiquitous protein in the hexosamine pathway which yields precursors required for protein and lipid glycosylation.

A GFPT1 knockout mouse model has been generated as part of the International Mouse Phenotyping Consortium. The GFPT1allele reports the activity of the promoter which we used to track the expression pattern of GFPT1 during development and across tissues. Since homozygous GFPT1 knockout mice are embryonic lethal, we bred and will characterise the conditional muscle-specific GFPT1 knockout mouse model. We will use in vitro isometric force measurements to assess muscle contractile function. In situ force measurements and the four limb inverted screen test will be used to measure fatigable muscle weakness. We will study the morphology of the synapse paying particular attention to the clustering properties of acetylcholine receptors and other proteins at the NMJ which are known to be glycosylated.

Heterozygous GFPT1 knockout mice display normal morphology of both the presynaptic and postsynaptic components at the NMJ. Furthermore, ?-galactosidase activity shows ubiquitous GFPT1 expression in both muscle and non-muscle tissues. Muscle-specific GFPT1 knockout mice are viable and normal in appearance and bodyweight. We show that any phenotype seen is due to a deficiency of GFPT1 in skeletal and cardiac muscle only. GFPT1deficient mice display lower hanging times when compared to wild type mice. This suggests that the muscle-specific GFPT1 knockout mouse demonstrates signs of muscle weakness.

GFPT1, glycosylation, neuromuscular junction

Congenital myasthenic syndromes- #2566

P19- 287- Are mutations of the skeletal muscle sodium channel Nav1.4 undoubtly associated with myasthenia, myopathy, and essential tremor?

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Nav1.4 is the main voltage-gated sodium channel expressed in adult skeletal muscles where it is crucial for generating and propagating the action potential. Mutations of SCN4A, which encodes the pore-forming subunit of Nav1.4, have been known to cause dominant non dystrophic myotonia and periodic paralyses since several years. Recessive SCN4A mutations have been reported only recently in 3 forms of muscle weakness: congenital myasthenia, f?tal hypokinesia, and congenital myopathy. The association of one heterozygous SCN4A variation (p.Gly1537Ser) to a dominantly inherited form of essential tremor has also been suggested, greatly complicating the pictures.

We will report 3 recessive variations of SCN4A, two (heteroallelic p.Val730Met and p.Gly1537Ser) in one patient suffering from a classical form of myasthenia and one (homozygous p.Arg1454Trp) in a patient presenting with a complex form combining myasthenia with periodic paralysis. Heterologous expression of mutant Nav1.4 excluded any major effect of p.Val730Met and p.Gly1537Ser on the channel activity in heterologous expression systems. Immunostaining of Nav1.4 suggested abnormal trafficking to the neuromuscular junction in the patient's muscle biopsy. These data question the relationship of p.Gly1537Ser with dominant essential tremor, as recently reported, as well as with congenital myasthenia in our patient. Important impairment of fast and slow inactivation compared to the wild type channel was recorded for p.Arg1454Trp. No major defect in cell trafficking at the neuromuscular junction was observed for this mutation compared to wild type when recombinant Nav1.4 were electroporated in adult skeletal muscles of rodents, which was found to be the most efficient way to experimentally investigate the Nav1.4 cell membrane trafficking.

All these investigations question the causative and pathophysiological relationship of recessive Nav 1.4 mutations with myasthenic and myopathic-like muscle weakness. Our data suggest that p. Gly1537Ser, reported to be associated with dominant essential tremor, could be a rare polymorphism. Altogether, with the development of whole exome sequencing, we recommend to perform full investigations on current density, gating behavior, and cell membrane trafficking at sarcolemma and at neuromuscular junction, for all newly observed SCN4A variant before concluding on its pathogenicity.

sodium channel, myasthenia, myopathy, periodic paralysis, essential tremor

Myasthenia gravis- #2429

P19- 288- The partial loss and irregular overexpression of caveolin-3 in muscle cell membrane is detected in myasthenia gravis patients

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Caveolin-3 is a muscle-specific membrane protein that localizes to the sarcolemma and T-tubule system and is critical for the correct functioning of skeletal muscle, repair of muscle membrane damage, and normal skeletal muscle development. Specific autoantibodies that destroy the post-neuromuscular junction underlie the pathogenesis of MG, and the mechanisms of neuromuscular junction restoration are not fully understood. Given the known functions of caveolin-3, we speculated that it acts to protect and repair the neuromuscular junction by inducing the differentiation of muscle cells and regulating sarcolemmal proteins. We therefore examined the expression of caveolin-3 in MG muscles to determine whether it is differentially expressed with respect to non-myogenic patient muscle.

We examined muscle biopsies from 15 of MG patients. Tissue samples were obtained from the musculus pectoralis major during thymectomy. In addition, five patients without myopathy provided control biopsy specimens from their upper or lower limb muscles. Immunohistochemistry, Western blot analysis and Quantitative real-time (qRT-) PCR analysis were carried out according to standard methods.

We observed that caveolin-3 is abnormally expressed in the muscle membrane of MG patients by immunohistochemistry, with partial loss of expression and overexpression observed in 5/15 and 10/15 patients, respectively. Caveolin-3 was overexpressed in MG muscle, specifically in small myofibers. The small myofibers likely represent regenerating myofibers arising from myoblast or myotube fusion, and the fact that caveolin-3 is present on these myofibers suggests that it plays a role in muscle repair.

Moreover, caveolin-3 protein expression was 2-fold higher in the muscle of MG as compared to non-myogenic patients (p = 0.008), as determined by western blot analysis. Similarly, a qRT-PCR analysis revealed that caveolin-3 mRNA expression was upregulated in MG relative to non-myogenic patients (p = 0.019).

The partial loss and irregular distribution of caveolin-3 in muscle cell membrane was detected in one-third of MG patients examined in this study suggests that this is a relatively common phenomenon in this disease; on the other hand, caveolin-3 upregulation may protect myotubes or restore the damaged neuromuscular junction. These findings provide insight into the molecular mechanisms underlying the cellular responses to muscle defects in MG.

myasthenia gravis, caveolin-3, muscle

P19- 289- Long-term outcome of juvenile myasthenia gravis. A nationwide population based study.

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Myasthenia gravis (MG) is an autoimmune disease targeting the neuromuscular synapsis, and affects children in 10-15 % of cases. The aim was to investigate the long-term outcome in a national population-based cohort of juvenile MG (JMG), to identify prognostic factors that could predict good outcome.
Patients and methods
Through systematic search in the electronic diagnostic systems at the 15 main hospitals in Norway, search in the AChR antibody database at Haukeland University Hospital and in the national MG database, we identified and included 63 patients with JMG, age of onset 1-17. The group was divided into pre- and post-pubertal onset, using age 12 as the arbitary cut of. Clinical data was collected by means of medical charts and in 51 patients an updated clinical evaluation. Symptoms were classified using Ossermann and MGFA scores, and outcome using MGFA Postintervention Status.
Results
Follow up ranged from 2 to 60 years. Mean age of onset was 11,9, and in 23 patients before age 12. There was a female preponderance, 55 girls and 8 boys. 92% had generalized MG and 74% were documented seropositive. 41 patients had undergone thymectomy, and all patients had utilized MG specific medication during the disease course. A significant share of all the patients did not require any drug in the long term and was in complete drug free stable remission (CSR).
Conclusion
Considering that a significant proportion of the patients evolve into complete remission, we wanted to identify prognostic factors that could predict good outcome. The preliminary data show that prepubertal onset MG might have a more favorable outcome, with a higher share of CSR, compared to postpubertal onset. Further results and analysis will be presented.

juvenile myasthenia gravis, long term outcome

P19- 290- MGEX: Myasthenia Gravis and EXercise, a randomised controlled trial.

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Introduction
Physical exercise (PE) has shown promising results in various neuromuscular diseases. To date, little research exists exploring the effects of PE in auto-immune myasthenia gravis (MG). The few existing studies present methodolgical flaws limiting the conclusions which can be drawn and the generalisability of results. We hypothesize that exercise could have potential positive physical and psychological effects as well as immunomodulatory effects on AIMG and may thus be a beneficial addition to the current therapeutic arsenal.
Objective
The aim of this study is to evaluate the feasability and demonstrate the benefits on perceived quality of life of a home-based physical exercise program for patients with stabilised, generalised auto-immune MG.
Methods
MGEX is a multi-centre, single blind, randomised controlled trial. Randomisation is performed by permuted blocks of varying sizes and stratified by center. Forty-two patients will be recruited. Age ? 18 ? 70 years old. The primary outcome measure is the change in the MGQOL-15-F score between 3 and 6 months. Other evaluations include the six-minute walk test, respiratory tests (FVC and MIP/MEP), the force and endurance of the Biceps Brachii and Vastus Lateralis via surface EMG as well as various psychological evaluations (WHO-QOL BREF, BECK, STAI, MINI, SEI). Habitual physical activity will be measured via accelometry. Following a 3 month period of observation patients will be randomised into either the control or treatment group. The treatment group will participate in a 40 minute physical exercise program using a rowing machine, 3 times a week for 3 months at their home. All patients will be followed up for a further 3 months.
Trial registration: ClinicalTrials.gov Identifier: NCT02066519
Results
This study is ongoing. Currently fourteen patients have been recruited since beginning in October 2014. Recrutement difficulties include geographical limitations (Ile de France), age limitations, time availability for patients currently working as well as lack of symptoms for a large majority of stabilised patients. To date no significant adverse events have been reported in relation to exercise in the experimental group.
Conclusion
This study intends to improve the current knowledge base concerning the possibility and efficacy of regular physical activity for patients with stable, auto-immune MG.

Auto-immune myasthenia gravis, physical exercise, quality of life

P19- 291- Cross-cultural adaptation and validation of the French version of the MG-QOL 15

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Introduction

Quality of life (QOL) questionnaires seek to establish an objective measure of a patients self-perceived well-being or satisfaction. The score can be used to measure disease evolution, treatment efficacy and the extent to which a patient is coping with their disease. In a chronic disease such as myasthenia gravis (MG) where symptoms fluctuate and the prognosis is uncertain, an evaluation of QOL is essential. Currently no such measure exists in French which is specific for MG.

Aim: The aim of this study was to translate, culturally adapt and evaluate the psychometric properties of the MGQOL-15 scale for use with patients in France.

Design: Prospective, validation study.

Method: The MG-QOL15 is a 15 item auto-questionnaire which is valid and reliable (ref). Translation and cross-cultural adaption were performed according to international recommendations (4, 5, 6). Patients completed the MG-QOL15-F during their out-patient appointment in two hospitals (Pitié Salpetriere, Paris and Raymond Poincaré, Garches) and the Myasthenic Muscle Score, the MG-ADL and the WHO-QOL BREF were completed when possible and recorded. Test-retest reliability was evaluatued with a 2 day interval (patients returned the 2nd questionnaire via the post) using an intra-class correlation coefficient. Spearman correlation coefficients were calculated for validity measures.

Results: A total of 125 patients (74 females) were included, mean age 52.9 ± 18.5 years. Internal consistency was excellent (Cronbach's alpha = 0.92) as was test-retest reliability (ICC = 0.92, 95% CI: 0.89, 0.94, n = 90). Concurrent validity was good for both the MMS (rho = -0.519, p >0.001, n = 91) and the MG-ADL score (rho = 0.623, p >0.001, n = 47). Correlations were strongest for overall QOL and general health score on the WHO-QOL BREF (rho = 0.622 p >0.001, n= 72) and the physical health domain (r = 0.667 p >0.001, n = 69).

Conclusion: The MG-QOL15-F is valid and reliable and is now available for use in clinical practice and research for French speaking patients. Further testing is planned to evaluate its responsiveness.

Quality of life, Auto-immune myasthenia gravis, patient reported outcome measure

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P19- 292- Pilot data from national registry of myasthenia (MyReg) in Czech republic

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Background and purpose

Myasthenia gravis belongs to orphan disease with prevalence varying from 15-200 per million depending on country. The patient registries belong to the core activities which can help us in planning of the effective health care, assessing standards of diagnosis and care, and answer the questions concerning on epidemiologic data. The new Czech registry (MyReg) for patients suffering from myasthenia gravis should gather and pool data from neuromuscular centres in Czech Republic.

Methods

The technology, the data collection, storage and backup and their analyses are provided by the Institute of Biostatistics and Analyses, Masaryk University, Brno, CR. On-line data collection is based on a TRIALDB system developed on Yale University, Connecticut, USA, which is widely used for this purpose.

Data transfer is encrypted and the system is designed to prevent their unauthorized use. Laws and regulations in CR require having an informed consent from all patients whose data are used in the registry. All claims for personal data protection were met. Data are stored on the central server on Masaryk University in Brno. Pilot data for MyReg were gathered from patients of Neuromuscular Centre of University Hospital Brno. Selected characteristics of the disease were analysed.

Results

208 patients from Neuromuscular centre Brno were gathered and analysed The mean age at onset for women and men was 50.1 and 62.5 years respectively. The M:F sex ratio was 1.3:1. The diagnosis was made in 86 % patients within the first year after first symptom. Most frequent initial symptoms were ocular, occurring in 68 % of patients. Generalized myasthenia was found in 75 % patients. Antibody positivity (ACHR) were found in 93 % and 80.4 % of patients with generalized and ocular form respectively. Thymoma was present in 13 % of all patients. In detail, thymoma was diagnosed in 25.9 % and 8 % of early onset (50 years or younger) and late onset myasthenia respectively.

Conclusions

Pilot data from Neuromuscular Centre University Hospital Brno show that the registry is user friendly and ready for use. Analysis of preliminary data discovered some distinctions with published data. In contrast with other studies we report male predominance, and higher average age of onset. Furthermore, thymoma was more frequent in early onset myasthenia.

P19- 293- Molecular hallmarks in myasthenia gravis

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Myasthenia gravis (MG) is a T-dependent B-cell mediated autoimmune disorder characterized by impaired neuromuscular transmission, resulting in muscular weakness and fatigability. In most of the patients (over 80%) the disease is caused by autoantibodies against the acetylcholine receptor (AChR). However, the exact immunological mechanisms underlying the autoimmune response are still unknown. We exploited total RNA sequencing (RNA-seq) approach to study the transcriptional profile in peripheral blood mononuclear cells (PBMCs) of AChR-MG patients versus healthy controls. We aim at identifying a molecular signature of AChR-MG leading to better understand the molecular basis of the disease. Transcriptional analysis combined with Ingenuity Pathway Analysis (IPA) revealed that 128 genes and 9 microRNA (miRNA) precursors were differentially expressed in AChR-MG patients compared to healthy controls. Functional annotation performed by IPA showed that 17% (22 out of 128) of the differentially expressed genes were associated with viral infections. Some of them were selected and validated by NanoString technology, thus, suggesting that a viral infection-associated transcriptional pattern is dysregulated in AChR-MG patients. Moreover, in order to validate RNA-seq data, by qPCR, we found 3 miRNAs (i.e. miR-612, miR-3651, miR-3654) significantly up-regulated in AChR-MG patients compared to healthy controls. In conclusion, our data suggest the presence of a molecular signature in PBMCs associated with AChR-MG.

Myasthenia gravis, RNA-sequencing

P19- 294- Experience of diagnostic and treatment of myasthenia gravis.

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Department of neurology named after S.N. Davidenkov of I.I.Mechnikov's North-West State Medical University has been studying myasthenia since 1945 and so far has gained experience of diagnostics and treatment of all age groups of patients with myasthenia and various forms of the disease. The study objective was more precise definition of diagnostic criteria and determination of therapy strategy for various forms of myasthenia on the basis of pathogenetic and clinical features of the disease, long-time follow-up observation.
We have examined 1149 myasthenia patients with various forms of the disease: generalized form- 863 (75,1%) patients, ocular- 162 (14,1 %), bulbar- 124 (10,8 %) at the age from 18 up to 78 years and duration of disease from six months up to 29 years. Among them there were 792 women (68,9 %), 357 men (31,1 %), average age of patients was 37,4 years. 4 groups of myasthenia diagnostic criteria were isolated: clinical, pharmacological, electroneuromyography and immunologic. Myasthenia diagnosis is doubtless if confirmed by 4 diagnostics criteria, authentic- in the presence of 3 of them, probable- when have 2 and doubtful- in the presence of 1 criterion of diagnostics. Treatment of myasthenia patients was carried out using basic AChE inhibitors therapy with selection of strictly individual daily dose depending on the form of the disease and type of the medicine. 492 (42,8%) patients with myasthenia have undergone thymectomy, other patients had drug pathogenetic therapy. Glucocorticosteroid therapy was carried out in 375 (32,6%) patients and has appeared effective in 82,3% of cases. For the main indications cytostatic drugs were prescribed (azathioprine, cyclosporine and cyclophosphan)- to 108 (9,3 %) patients. Complex treatment of myasthenia was based on efferent therapy: exchange plasmapheresis, hemosorption and enterosorption with positive therapeutic effect accordingly in 89,6%, 86,7% and 61,2% cases.
Correct diagnostics and timely prescription of adequate methods of treatment result in compensation of motor disorders in more than 80,0% of patients. Selection of myasthenia treatment method can be defined by a disease form, severity level and pathogenic pathway. Future of scientific researches in patients with myasthenia is related to more detailed study of pathogenetic mechanisms and use of targeted immunotherapy which has specific influence on the main effector molecules- main components of pathogenesis.

Myasthenia gravis, diagnostic, treatment

P19- 295- Extracorporeal photochemotherapy in the treatment of myasthenia gravis

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Objective: to evaluate to efficacy of extracorporeal photochemotherapy (EKFHT) in patients with myasthenia gravis.
Material and methods: the treatment was carried out on 31 adult patients with generalized myasthenia outside he period of exacerbation. Muscle strength was evaluated by 5 point sistem. Method EKFHT was as follows: patients were subjected to discontinuous leukapheresis procedure, during wich 1 liter of blood was removed aproximately 200-250 ml of leukocyte enriched blood containing an averidge 2,43-3,6x10*9 mononuclear cells. Befor reinfusion leukocites irradiated with ultraviolet (UV). The course consisted of 4 sessions (2 times a week with for 2 sessions). Befor each session the patient was given the photosensitizing medicine. Conduct research subpopulation of lymphocytes, interferon status, immunoglobulins.
 Results: in 27 of 31 patients (87%) showed improvement. Significantly increased the power of facial muscles, neck muscles, biceps, deltoid, iliopsoas and gastrocnemius muscles (p>0,05). The clinical effect lasted up to 3-4 months. There up to 2 years of observation, when the patient is admitted to the bulbar and oculomotor disturbanses. Later in the regular conduct of the

tratment of bulbar disorders were no more, and with the appearence light ghosting after 1 session is held. Lymphocyte subpopulation mainly decreased, normolized immunoregulatory index CD4/CD8. Decreased concentration of immunoglobulins, mainly IgA(18%), less IgG 98,3%) and IgM (2%). The average values of interferon status decreased, but not statistically significant.
Conclusion: EKFHT- effective and promising method for the treatment of myasthenia gravis. EKFHT- an alternative method of treatment of the disease, which does require the use of glucocorticoids and cytoctatics

myasthenia, photochemotherapy

Myasthenia gravis- #2900
P19- 296- Clinical follow-up of the pregnancy in myasthenia gravis patients.

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Introduction. Myasthenia gravis (MG) has highest incidence in the third decade of the women's life which overlapping with the childbearing years, therefore it is important to known if pregnancy itself has some impact in the course of MG and if MG has some impact to pregnancy as well.
Objectives: The aim of this study was analyse the outcome and course of disease and pregnancy in MG patients.
Methods: We analysed retrospectively MG women who have pregnancy from 1990 to 2015. Obstetric and clinical data were analysed before, during and after pregnancy.
Results: We included 37 pregnancies from 23 MG patients. Obstetric complications were reported in 23 pregnancies. The preterm rupture of amniotic membranes was the most common complication (10 premature births) and severe complication occurred in 5 patients. The mode of delivery was caesarean section in 21 women, vaginal with forceps in 2 and spontaneous vaginal delivery in 10. Among 30 pregnancies, the MG symptoms improved in 9 pregnancies, remained unchanged in 6 and worsened in 15. Second trimester and postpartum were the main period for worsening the MG symptoms. Twenty-two patients received spinal anaesthesia, two patients received local anaesthesia and there were no complications. In MG therapy, only 3 patients were without any therapy before pregnancy and 27 patients were treated using pyrydostigmine and immunosuppressive drugs. Neonatal myasthenia gravis occurred in 4 babies from 33 babies lived born.
Conclusion. The worsening of the MG symptoms occurs more in the second trimester and postpartum. MG course in former pregnancy does not predict the course of following pregnancies. MG pregnant has increasing rates of the premature labour by preterm rupture of amniotic membranes and caesarean delivery by obstetric indication. The anaesthesia can be safety recommended. However, the pregnancy factors that could influence clinical severity in the MG still are unpredictable.

myasthenia gravis, pregnancy, pregnancy complications, management, anaesthesia.

Myasthenia gravis- #2950
P19- 297- FUNCTIONAL ALTERATION OF SATELLITE CELLS IN MYASTHENIA GRAVIS: KEY ROLE OF MYOD AND MYOG

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Myasthenia gravis (MG) is a neuromuscular disease caused by autoantibodies against Acetylcholine Receptor. MG is characterized by fatigability and fluctuating muscle weakness. Muscle homeostasis and regeneration is carried out by local stem cells called satellite cells (SCs). However, molecular and cellular mechanisms of myogenesis in MG are still unknown.
Muscle biopsies from MG and healthy age-matched controls were collected. SCs were isolated using explants culture and positive selection of CD56+ cells. Proliferation and differentiation of myoblasts in vitro were assessed by cell counting and muscle myosin immunolabeling (from day 0 to day 4). Localization of myogenic markers in muscle biopsies of MG patients and healthy controls was determined using immunolabeling. mRNA expression was analysed by real-time qPCR.
We show that SCs from MG muscle proliferated and differentiated more actively than SCs from healthy muscles. During these processes, MyoD and MyoG were expressed at a higher level in MG SCs. Additionally, MyoD and MyoG also expressed at a higher level in MG muscle biopsies. These results suggest that MyoD and MyoG could be responsible for the functional differences observed in SCs from MG compared to healthy muscles.
We also show that SCs from healthy muscles and treated with MG sera or monoclonal AChR antibodies differentiated more and expressed more MyoG mRNA than those treated with Ctrl sera or isotype antibodies. These results suggest that increased differentiation and MyoG expression could be due to the anti-AChR antibodies present in the MG sera. Therefore, AChR antibodies could play a key role in the SCs differentiation via the modulation of MyoG expression.
Altogether, these findings demonstrate that the autoimmune attack in MG might lead to important changes in the function of SCs that could represent a mechanism of compensation to regenerate muscle fibres that have been damaged by the autoantibodies or maintain muscle mass.

Satellite cells, Myasthenia gravis, functional alterations

Myasthenia gravis- #3177
P19- 298- Rituximab for the treatment of pediatric autoimmune neuromuscular disorders

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Myasthenia gravis (MG) and chronic inflammatory demyelinating polyneuropathy (CIDP) are autoimmune neuromuscular disorders affecting both adult and children. There is currently very little data on the use of newer immunomodulatory agents, such as Rituximab (RTX), in these children.
Objective: To describe our single-center experience with RTX for the treatment of pediatric patients with MG or CIDp.
Methods: Retrospective chart review of all pediatric patients with MG or CIDP who received RTX at our institution since 2008. Clinical presentation, age at diagnosis, investigations, serological profile, medications, hospitalizations, time from diagnosis to RTX, pre- and post-treatment modified Rankin scale (mRS), pre-treatment Myasthenia Gravis Foundation of America (MGFA) class and postintervention status, adverse effects and follow-up length were recorded.
Results: We identified four pediatric patients with MG and two with CIDP who received RTX. Mean age at diagnosis was 8 years (range: 2-14). Average time from diagnosis to RTX initiation was 27 months (range: 8-43). Average length of follow-up after RTX was 37 months (range: 14-68). At the latest follow-up, four patients had improved compared to their pre-RTX baseline mRS status. Two patients, both of whom had MG, were clinically unchanged. No patient achieved complete remission and all six patients were still on at least one immunomodulatory treatment other than RTX. No adverse effect of RTX was noted.
Discussion: In four out of six patients, RTX therapy led to clinical improvement, but not to complete remission. These four patients achieved sustained clinical improvement after a single RTX course, whereas the other two had no significant change in clinical status after several RTX courses.
Conclusion: A single course of RTX can result in significant and sustained clinical improvement for some pediatric patients with MG or CIDP, but larger studies are needed to determine which patients would benefit most.

chronic inflammatory demyelinating polyneuropathy, rituximab, pediatric autoimmune neuromuscular disorders

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P20 –Myotonic syndromes (dystrophic and non-dystrophic)- N° 299 to N° 322

Myotonias (except DM1 and DM2)- #2278
P20- 299- Electromyography in diagnosis of hereditary myotonic syndromes

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Hereditary myotonic syndromes (HMS)-a small genetically heterogeneous group of a muscle channelopathies with marked clinical polymorphism and often overlapping phenotypes. Myotonia is the leading symptom of HMS, is a result of the membrane excitability of muscle fibers, whose functional consequences can be studied by EMG. The correct EMG analysis can help in diagnosis of different types Myotonic dystrophy (DM), Nondystrophic myotonia (NDM) and to predict possible HMS causative mutations.
EMG was conducted in 88 DNA detected patients with HMS (Becker's disease (BD), Thomsen's disease (TD), DM type 1(DM1) and 2(DM2)). Repetitive nerves stimulation (RNS) 4/s at a rate of 50 Hz (RNS1) was done 64 patients, 57 patients of 30 Hz (RNS2), 58 patients of 10 Hz (RNS3). SET was held in 58 patients according to Fournier I, II, III patterns. 87 patients had needle EMG with an average duration of 3 typical myotonic discharge (MyD) (Table).
CF. Figure
We found no significant statistical differences in the values of the EMG in the patients with ?D and BD, similarly in the patients with DM1 and DM2. RNS2 and RNS3 were not enough in proving in diagnostic our patients' HMS.

The most significant changes were received in NDM (TD and BD) and DM (DM1 and DM2). The CMAP decrement was less than 60% in RNS1 in 18(78.3%) DM patients and only 5(16.1%) NDM patients. In NDM patients the decrement remained stable for over 5 years. One patient with homozygous CLCN1 gene mutations c.1936A>G did not have decrement in RNS1. MyD were longer than 1.5 seconds in 35(83.3%) DM patients, and only 4(9.5%) NDM patients, 2 DM1 patients did not have MyD.
Pattern II in SET was detected only 5(20.8%) NDM patients. Pattern III in SET was found in 42(80.8%) NDM and DM patients.
RNS1 and MyD allows to distinguish between NDM and DM patients (p>0,01) and the stable persistent CMAP decrement in the NDM will be possibly important in predicting of the individual mutations in the CLCN1 gene. The fact that Fournier's pattern II in SET was not informative in TD and BD patients needs to be explained in further researches.
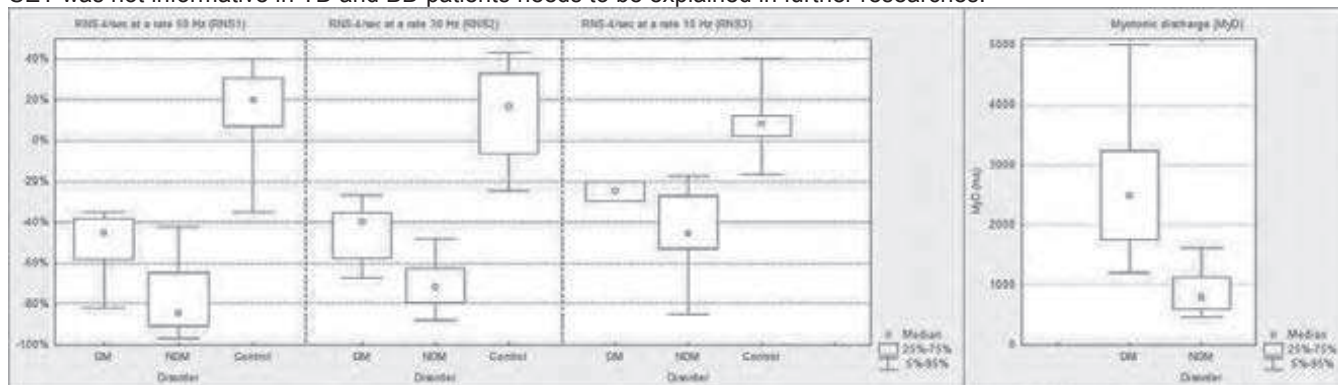
Becker's disease, Thomsen's disease, EMG, repetitive nerves stimulation, myotonic discharge, short exercise test, channelopathies