

**PW 23:**  
**Hereditary neuropathies**

PW23-280	<p><b>PATHOPHYSIOLOGICAL MECHANISMS IN LAMIN A/C ASSOCIATED CHARCOT-MARIE-TOOTH DISEASE (CMT2B1)</b></p> <p>POITELON Y<sup>1</sup>, BAUDOT C<sup>1</sup>, HAMADOUCHE T<sup>1</sup>, LEVY N<sup>1</sup>, DELAGUE V<sup>1</sup>  (1) Inserm UMR_S 910, Génétique Médicale &amp; Génomique Fonctionnelle, Faculté de Médecine de la Timone, Marseille, FRANCE.</p>
<p>To contact the author::  Yannick.Poitelon@univm  ed.fr.</p>	<p>Lamins, a class of intermediate filaments, are major components of the nuclear lamina, a filamentous network underlying the inner face of the nuclear membrane. A-type Lamins are encoded by the same gene: <i>LMNA</i>, regulated by the AP1 complex (c-Fos &amp; c-Jun). Up to date, eleven pathologies, described as laminopathies, have been associated with mutations in <i>LMNA</i>. One of these, Charcot-Marie-Tooth disease (CMT), type 2B1, is an autosomal recessive form of axonal CMT caused by the c.892C&gt;T transition in <i>LMNA</i> exon 5. (p.Arg298Cys). In order to progress towards understanding of the pathophysiological mechanisms underlying CMT2B1, we studied two different models for the disease: human cells from patients homozygous for the c.892C&gt;T mutation, and a Knock-In <i>Lmna</i><sup>R298C/R298C</sup> mouse model.</p> <p>Gene expression studies performed on human microfluidic plates (Low Density Arrays) evidenced significant decrease in expression levels of several genes, including <i>LMNA</i>. These observations were confirmed in mouse brain, skeletal and cardiac muscle, sciatic nerve and spinal cord at the transcriptional level, as well as on lymphoblastoid human cell lines at the protein level.</p> <p>The p.Arg298Cys mutation lies within a coil-coiled domain, an important functional domain for intermediate filament polymerization. In silico predictions are in favor of a potential destabilizing effect of the mutation. Moreover, previous publications have shown that Lamins interact with c-Fos, a Leucine Zipper transcription factor, through their coiled-coil domain. We therefore propose a two-hit pathophysiological mechanism model:</p> <ul style="list-style-type: none"> <li>- The pArg298Cys mutation might destabilize complexes between A-type Lamins and transcription factors: the latter might be either components of the AP1 complex, or some nerve specific transcription factors, which remain to be identified.</li> <li>- <i>LMNA</i> seems to be autoregulated through an A_type Lamins – AP1 complex, which might be disrupted at the DNA level by the presence of the mutation.</li> </ul> <p>We are actually conducting further experiments in order to confirm these hypotheses.</p>

PW23-281	<p><b><u>ABNORMAL INTERACTION OF MUTANT HSP22 (HSPB8) WITH THE RNA HELICASE DDX20 (GEMIN3, DP103) IN DISTAL HEREDITARY MOTOR NEUROPATHY AND CHARCOT-MARIE-TOOTH DISEASE.</u></b></p> <p>SUN X<sup>1</sup>, FONTAINE JM<sup>1</sup>, SIMON S<sup>2</sup>, HOPPE A<sup>1</sup>, CARRA S<sup>3</sup>, DE GUZMAN C<sup>1</sup>, MARTIN J<sup>4</sup>, VICART P<sup>5</sup>, LANDRY J<sup>6</sup>, WELSH M<sup>1</sup>, BENNDORF R<sup>7</sup></p> <p>(1) University of Michigan, Ann Arbor, USA. (2) CGMC-UMR5534-Stress, Chaperons et Mort Cellulaire, Lyon, FRANCE. (3) University Medical Center Groningen, Groningen, NETHERLANDS ANTILLES. (4) Loyola University Chicago, Maywood, USA. (5) Université Paris 7 - EA300, Paris, FRANCE. (6) Université de Laval, Québec, CANADA. (7) Center for Clinical and Translational Research, Columbus, USA.</p>
To contact the author:: stef.labo@gmail.com.	<p><b>Eight mutations in the small heat shock proteins (sHSP) Hsp22 and Hsp27 have been associated with the motor neuron diseases (MND) distal hereditary motor neuropathy and Charcot-Marie-Tooth disease. Hsp22 and Hsp27 interact with each other, suggesting that these two etiologic factors may act in the same pathway. In an effort to learn about the role of Hsp22 in MND, we screened a human cDNA library by the yeast two-hybrid method for potential binding proteins. One identified protein was the RNA helicase Ddx20, a core component of the survival-of-motor neuron (SMN) complexes. This interaction was verified by independent methods including FRET. Both mutant Hsp22 forms showed abnormally increased binding to Ddx20. Interestingly, Ddx20 itself binds to the SMN protein, and mutations in the <i>SMN1</i> gene cause spinal muscular atrophy, another MND. Thus, these protein interaction data have linked the etiologic factors Hsp22, Hsp27, and SMN, and mutations in any of these genes cause the various forms of MND. SMN complexes are involved in RNP processing. The mutant Hsp22/Ddx20 interaction was sensitive to treatment with RNase suggesting involvement of RNA in this interaction and a potential role of sHSPs in RNP processing.</b></p>

PW23-282	<p><b><u>MOLECULAR EXPLORATION OF AXONAL CMT IN A LARGE SERIES OF PATIENTS : FOCUS ON MFN2 AND GDAP1 GENES.</u></b></p> <p>BONELLO-PALOT N<sup>1</sup>, LATOUR P<sup>2</sup>, MARTINI N<sup>1</sup>, MAYENÇON M<sup>2</sup>, PÉCHEUX C<sup>1</sup>, MÉGARBANÉ A<sup>3</sup>, ATTARIAN S<sup>4</sup>, POUGET J<sup>4</sup>, LÉVY N<sup>5</sup>, BERNARD R<sup>1</sup></p> <p>(1) Département de Génétique médicale Laboratoire de Biologie Moléculaire CHU Timone, Marseille, FRANCE. (2) Neurogénétique Centre de Biologie Groupement Hospitalier Est CHU, Lyon, FRANCE. (3) Unité de Génétique Médicale, Faculté de Médecine, Université Saint-Joseph de Beyrouth, Paris, FRANCE. (4) Service des maladies neuromusculaires CHU Timone, Marseille, FRANCE. (5) Inserm UMR 910 Faculté de Médecine, Marseille, FRANCE.</p>
To contact the author:: nathalie.bonello-pallot@ap-hm.fr.	<p>Charcot-Marie-Tooth neuropathies (CMT), also known as hereditary motor and sensory neuropathies (HMSN), are a group of genetically and clinically heterogeneous diseases of the peripheral nervous system. 40 genes and more than 60 loci have been identified to date. The French CMT diagnosis network has defined 6 types of strategies for molecular exploration, linked to 6 phenotypic sub-types of CMT. We focused our study on type 2 CMT (axonal) which phenotypic presentation is characterised by relative preservation of nerve conduction velocity (&gt;38m/s in the median nerve), with decrease in compound motor action potentials providing evidence of axonal loss. We studied in particular <i>MFN2</i> (Mitofusine 2) and <i>GDAP1</i> (Ganglioside Induced Differentiation-Associated Protein 1) genes because of several points of particular interest. First, their mutational spectrum is large and only recently described, making molecular interpretation of news variants and subsequent genetic counselling difficult. Second they are both implicated in mitochondrial dynamic. Third their place within the exploration strategy diagram is not completely solved to date. We aimed to estimate the frequency of mutations in <i>MFN2</i> and <i>GDAP1</i> genes in a big cohort of CMT patients collected in France and to evaluate the potential pathogenicity of new mutations. The frequency of mutation in <i>MFN2</i> gene is about 11% (25/226) and in <i>GDAP1</i> gene is about 6.7% (9/135) in our study. Indeed, 75% (6/8) of <i>GDAP1</i> sequence variations and 72% (18/25) of <i>MFN2</i> sequence variations, we found weren't reported to date and raised to a lot of difficulties in genetic counselling. We report in particular and discuss a missense mutation in <i>GDAP1</i> that seems to segregate with the disease in both dominant and recessive mode of inheritance.</p>

PW23-283	<p><b><u>FGD4, ENCODING THE RHOGEF FRABIN, IS IMPLICATED IN CHARCOT MARIE TOOTH TYPE 4H</u></b></p> <p>BAUDOT C<sup>1</sup>, POITELON Y<sup>1</sup>, HAMADOUCHE T<sup>1</sup>, JACQUIER A<sup>2</sup>, BOCCACCIO I<sup>1</sup>, CHOUERY E<sup>3</sup>, CHAOUCH M<sup>4</sup>, KASSOURI N<sup>4</sup>, JABBOUR R<sup>5</sup>, GRID D<sup>6</sup>, MÉGARBANÉ A<sup>3</sup>, HAASE G<sup>2</sup>, LÉVY N<sup>1</sup>, DELAGUE V<sup>1</sup></p> <p>(1) U 910 Génétique Médicale et Génomique Fonctionnelle, Marseille, FRANCE. (2) IBDM, Marseille, FRANCE. (3) Université Saint-Joseph, Beirut, LEBANON. (4) Centre Hospitalier Universitaire Ben Aknoun, Alger, ALGERIA. (5) American University of Beirut Medical Center, Beirut, LEBANON. (6) Généthon III, Evry, FRANCE.</p>
<p>To contact the author:: cecile.baudot@univmed.fr.</p>	<p>Charcot-Marie-Tooth (CMT) disorders are a clinically and genetically heterogeneous group of hereditary neuropathies characterized by chronic distal weakness and sensory loss. CMT4H is an autosomal recessive demyelinating subtype recently mapped by us at chromosome 12p11.21-q13.11, in two consanguineous families of Mediterranean origin. In both families, we identified mutations in <i>FGD4</i>, encoding FGD4/FRABIN, a Rho GDP/GTP Exchange Factor (Rho GEF) specific to RhoGTPase Cdc42. Both mutations affect the same codon: the p.Met298fsX8 (c.893T&gt;G) in the Lebanese and the p.Met298Thr (c.893T&gt;C) in the Algerians. When overexpressing wild-type and truncated (p.Met298fsX8) forms of Frabin in rat primary motoneurons and rat RT4 Schwannomacells, we observed that the truncated form of Frabin induced significantly fewer microspikes than the wild-type. At the transcriptional level, we showed a 60% reduction in <i>FGD4</i> mRNA levels in Lebanese patient's fibroblasts, indicating that the truncated mRNA might be degraded by NMD. Moreover, a broad transcriptional study in different human tissues led us to characterize 17 alternative transcripts for <i>FGD4</i>. Interestingly, some of them are deleted of several exons and the corresponding protein lacks one or more functional domains. In consequence, we propose that FRABIN might have different roles in different tissues, depending on the functional domains present on the protein.</p> <p>In conclusion, FRABIN is the first RhoGEF to be identified in CMT disease. Several mechanisms and pathways leading to the pathology remain to be elucidated. However, three main hypothesis might be proposed: 1) loss of Cdc42/Rac1 activation, leading to a disorganization of the cytoskeleton, and perturbation of movements and/or migration 2) disruption of JNK pathway, affecting myelination process 3) implication of FRABIN in the activation of mitochondrial GTPases mutated in CMT (MFN2...), and perturbation of mitochondrial dynamics.</p> <p>We show here some primary results, further experiments are under process in order to validate our hypothesis.</p>

PW23-284	<p><b><u>A NOVEL PERIAXIN MUTATION CAUSES LATE ONSET AND SLOW PROGRESSIVE CHARCOT-MARIE-TOOTH DISEASE</u></b></p> <p>NOUIOUA S<sup>1</sup>, BERNARD R<sup>2</sup>, HAMADOUCHE T<sup>3</sup>, VALLAT JM<sup>4</sup>, LEVY N<sup>2</sup>, TAZIR M<sup>1</sup>  (1) Service de Neurologie, CHU Mustapha, Algiers, ALGERIA. (2) Département de Génétique Médicale, Laboratoire de Génétique Moléculaire, Hôpital d'Enfants de la Timone, Marseille, FRANCE. (3) Laboratoire de Biologie Moléculaire, Institut Pasteur, Algiers, ALGERIA. (4) Service de Neuropathologie, Hôpital Dupuytren, Limoges, FRANCE.</p>
<p>To contact the author::  merientazir@yahoo.com</p>	<p>Autosomal recessive forms of Charcot-Marie –Tooth disease are clinically and genetically heterogeneous. One locus, termed CMT4F, showing similarities to Déjerine- Sottas syndrome, was mapped to 19q13.3 in a large consanguineous Lebanese family and a mutation in the periaxin gene (<i>PRX</i>) was identified in this family. In the recent years, only seven other mutations were described in this gene. Here, we report a large family with four adolescent and adult patients harbouring a novel homozygous c.1090C&gt;T (p. Arg364Stop) mutation in exon 7 of the <i>PRX</i> gene. The clinical phenotype is characterized by a relatively late onset demyelinating sensory motor neuropathy with spine deformities (kyphoscoliosis) and feet deformities. Sensory ataxia was also present in the 2 youngest patients. In contrast with the majority of the CMT4F patients already reported, evolution is slowly progressive and the 4 patients have a mild motor disability. Nerve biopsy shows severe loss of myelinated and unmyelinated fibers and some myelin outfoldings. This is the first Algerian CMT4F family reported, and the causative <i>PRX</i> mutation is responsible for a moderate CMT phenotype with some intrafamilial variability.</p>

PW23-285	<p><b>PHENOTYPICAL FEATURES OF 14 MOROCCAN FAMILIES WITH AUTOSOMAL RECESSIVE CHARCOT-MARIE-TOOTH DISEASE ASSOCIATED WITH MUTATIONS IN THE GDAP1 GENE</b></p> <p>BIROUK N<sup>1</sup>, BOUHOUCHE A<sup>2</sup>, BELAÏDI H<sup>1</sup>, BENOMAR A<sup>2</sup>, AZZEDDINE H<sup>3</sup>, DUBOURG O<sup>4</sup>, MAISONOBE T<sup>4</sup>, YAHYAOUÏ M<sup>2</sup>, LE GUERN E<sup>3</sup>, OUAZZANI R<sup>1</sup></p> <p>(1) Service de Neurophysiologie Clinique, Rabat, MOROCCO. (2) Service de Neurologie et Neurogénétique, Rabat, MOROCCO. (3) INSERM U289, Hôpital de la Salpêtrière, Paris, FRANCE. (4) Laboratoire de Neuropathologie Raymond Escourrolle, Hôpital de la Salpêtrière, Paris, FRANCE.</p>
To contact the author:: birna@menara.ma.	<p>Mutations in GDAP1 gene located in 8q13 chromosome have been identified in families with either axonal or demyelinating form of autosomal recessive Charcot-Marie-Tooth (CMT) disease. Twenty five patients belonging to 14 Moroccan consanguineous families were examined clinically and electrophysiologically. In one patient, a peroneal nerve biopsy was performed. Linkage to 8q13 was then demonstrated and a mutation in the coding region of the GDAP1 gene was identified by direct sequencing.</p> <p>Neuropathy was evident during early childhood, walking was delayed in 5 cases and onset of symptoms occurred before 6 years in the others. The phenotype was very severe: foot deformities and disability involving the hands and feet developed towards the end of the first decade and followed by involvement of proximal muscles in the lower limbs leading to loss of autonomy in 16 cases. Only three patients had hoarse voice. Thirteen patients belonging to 7 families were homozygous for the S194X mutation of GDAP1. They all had axonal form of CMT according to electrophysiological and morphological findings. Six patients belonging to 3 families had either homozygous P78L mutation or compound heterozygous P78L/S194X mutations. Most of them had markedly reduced motor nerve conduction velocity ranging from 11 to 34 m/s consisting with a demyelinating form of CMT. For the remaining 6 patients belonging to 3 families, GDAP1 mutation has not been identified yet.</p> <p>The main phenotype characteristics of GDAP1 mutations are: early onset in childhood, severity and important foot deformities. The homozygous S194X mutation is the most frequent in our families and was related to axonal form of CMT suggesting a probable founder effect and a possible phenotype/genotype correlation.</p>

PW23-286	<p><b><u>GAIT ANALYSIS IN CHARCOT MARIE TOOTH (CMT) DISEASE: PRELIMINARY STUDY</u></b>  BOULAY C<sup>1</sup>, POMERO V<sup>2</sup>, JACQUEMIER M<sup>2</sup>, CUSTAUD M<sup>3</sup>, PAGNI S<sup>2</sup>, VIEHWEGER E<sup>2</sup>, GLARD Y<sup>2</sup>, JACOPIN S<sup>2</sup>, LAUNAY F<sup>2</sup>, JOUVE JL<sup>2</sup>, BOLLINI G<sup>2</sup>, CHABROL B<sup>1</sup>  (1) Centre de Référence des Maladies neuromusculaires de l'enfant (Pr. Chabrol), CHU Timone enfants, Marseille, FRANCE. (2) Laboratoire d'analyse du mouvement, service de chirurgie orthopédique pédiatrique (Pr. Bollini), CHU Timone enfants, Marseille, FRANCE. (3) Lecante Orthosud, Marseille, FRANCE.</p>
To contact the author:: christophe.boulay@ap-hm.fr.	<p>The gait analysis provided different kinematic patterns of walking in the Charcot Marie Tooth disease (CMT). The dynamic electromyography (EMG) described the muscles pattern for each type of gait in CMT.</p> <p>The classical gait pattern in CMT is characterized by a drop foot during the swing phase: the dorsal flexor muscles of ankle were weak in relation to the plantar flexor muscles. But the drop foot was often absent and in this case it was the plantar flexor muscles of ankle which were weak in relation to the dorsal flexor muscles. Also the gait analysis described a delayed and/or increased peak of dorsal flexor ankle, during the stance phase: the heel lift ("third rocker") is delayed. These data are studied in a sample of 10 subjects.</p> <p>Thus the dynamic EMG revealed the balance between agonist and antagonist ankle muscles and the reliability of the gait pattern and the foot deformity. It performs the importance of this functional exam. These neurophysiological and dynamic data facilitate the therapeutic strategy (orthosis, physical therapy, surgery) and the fonctionnal evaluation.</p>

PW23-287	<p><b>PHENOTYPIC VARIABILITY IN GIANT AXONAL NEUROPATHY</b>  NOUIOUA S<sup>1</sup>, MAGY L<sup>2</sup>, HAMADOUCHE T<sup>3</sup>, RAUTENSTRAUSS B<sup>4</sup>, GRID D<sup>5</sup>, ASSAMI S<sup>6</sup>, VALLAT JM<sup>7</sup>, TAZIR M<sup>8</sup></p> <p>(1) Service de Neurologie, CHU Mustapha, Algiers, ALGERIA. (2) Service de Neuropathologie, Hôpital Dupuytren, Limoges, FRANCE. (3) Laboratoire de Biologie Moléculaire, Institut Pasteur d'Algérie, Algiers, ALGERIA. (4) MGZ-Medizinisch Genetisches Zentrum, München, GERMANY. (5) Genethon, Evry, FRANCE. (6) Service de Neurologie, CHU Mustapha, Algiers, ALGERIA. (7) Service de Neuropathologie, Hôpital Dupuytren, Limoges, FRANCE. (8) Service de Neurologie, CHU Mustapha, Algiers, ALGERIA.</p>
To contact the author:: meriem.tazir@gmail.com	<p>Giant axonal neuropathy (GAN) is a recessively inherited neurological disorder affecting both central and peripheral nervous system. The main pathological hallmark of the condition is abnormal accumulation of intermediate filaments in giant axons and other cell types. Mutations in the <i>GAN</i> gene encoding Gigaxonin are responsible for the phenotype.</p> <p>We report clinicopathological, neurophysiological and genetic data from 15 patients belonging to 7 families with giant axons observed on nerve biopsy.</p> <p>In 6 families, we identified 3 different disease-causing homozygous mutations in the <i>GAN</i> gene. The c.1429C&gt;T (R477X) mutation which seems prevalent, was observed in 4 unrelated families originating from eastern Algeria. This mutation was responsible for 3 different phenotypes: 5 patients from 2 families had the classical GAN clinical picture with kinky red-hair, sensory motor axonal neuropathy, moderate mental retardation and cerebellar syndrome. The third family patient had congenital neuropathy with arthrogyriposis and the fourth family patient had a pyramidal paraparesia with facial diplegia. All the patients of these 4 families had a diffuse cerebral dysmyelination on MRI.</p> <p>The 2 other families linked to the <i>GAN</i> gene had a CMT-like presentation. One family with 4 patients had a c. 431G&gt;A (R138H) mutation, and the second family patient had a c.505G&gt;A (E169K) mutation. In the seventh family also with a CMT-like phenotype but no mutation in the <i>GAN</i> gene, we identified a c.2710C&gt;T (R904X) mutation in the <i>SH3TC2</i> gene implicated in CMT4C.</p> <p>In conclusion, the phenotypic consequences of <i>GAN</i> mutations can be variable. The same mutation c.1429C&gt;T (R477X), found in 4 families from the same geographical area suggesting a founder effect, is responsible for different phenotypes. CMT-like presentation with characteristic giant axons may be linked to mutations in the <i>GAN</i> gene but also in other genes, especially the <i>SH3TC2</i> gene (CMT4C) and the <i>NEFL</i> gene (CMT2E).</p>

PW23-288	<p><b><u>CELLULAR MODELS FOR GIANT AXONAL NEUROPATHY: DEVELOPMENT OF A NEW DIAGNOSTIC TOOL AND ASSESSMENT OF THE ROLE OF TUBULIN CHAPERONES IN VIMENTIN AGGREGATION</u></b></p> <p>CLEVELAND D<sup>1</sup>, YAMANAKA K<sup>2</sup>, BOMONT P<sup>3</sup></p> <p>(1) Ludwig Institute for Cancer Research, University of California San Diego, La Jolla, USA. (2) RIKEN Brain Science Institute, Saitama, JAPAN. (3) INMED - Inserm U901; Aix Marseille Université, Marseille, FRANCE.</p>
To contact the author:: pbom@inmed.univ-mrs.fr.	<p>Mutations in the gene encoding gigaxonin are causative for the fatal, early-onset recessive neurodegenerative disorder Giant Axonal Neuropathy (GAN). The crucial role of gigaxonin in neuronal maintenance, first assessed by alterations in the motor/sensory tracts of the peripheral nervous system and the impairment of the central nervous systems was subsequently confirmed in the GAN mouse model. Characterized by a generalized aggregation of Intermediate Filaments (IFs), GAN points to the essential role of cytoskeleton architecture in neuronal function, and especially to the implication of IF disorganization in neurodegeneration. Identification of gigaxonin as the substrate adaptor of a new Cul3-ubiquitin ligase E3 and three of its partners (the microtubule associated proteins MAP1B, MAP8 and the tubulin chaperone TBCB) allows now to dissect the mechanisms of neurodegeneration and IFs organization in GAN.</p> <p>We studied here cellular models of GAN A) to develop an alternative diagnostic tool (to nerve biopsies) for GAN patients and B) to assess the role of microtubules (MTS) and tubulin chaperones in IF aggregation.</p> <p>A) Using our new gigaxonin specific monoclonal antibodies and protein extracts from lymphoblast cell lines derived from GAN patients, multiple disease causing mutants are shown to be unstable, demonstrating that GAN is caused by the loss of function of gigaxonin.</p> <p>B) We assessed the implication of MTs and tubulin chaperones in vimentin organization in transfection experiments but also in the GAN cellular model: the patients's skin derived primary fibroblasts. This allowed us to demonstrate that neither MTs instability nor TBCB overabundance are able to reproduce the vimentin aggregates so characteristic of GAN patients.</p>

PW23-289	<p><b><u>AHNAK EXPRESSION BY MYELINATING SCHWANN CELLS REVEALS CAJAL BANDS IN VIVO AND IS INVOLVED IN THEIR MORPHOGENESIS AND LAMININ SUBSTRATE ADHESION IN VITRO</u></b></p> <p>VON BOXBERG Y<sup>1</sup>, SALIM C<sup>1</sup>, ALTERIO J<sup>1</sup>, FÉREOL S<sup>1</sup>, NOTHIAS F<sup>1</sup>  (1) UPMC Paris06-CNRS UMR 7101 "NSI", Paris, FRANCE.</p>
To contact the author:: ysander.boxberg@snv.jussieu.fr.	<p>Relatively little is known about the precise molecular mechanisms regulating the differentiation-associated morphological changes during Schwann cell (SC) development and regeneration/remyelination after injury, implying specific interactions with the local environment that ultimately converge on a reorganization of the cytoskeleton. We investigated the role in SC differentiation and function of the giant phosphoprotein AHNAK, which we show for the first time to be expressed at high levels in developing and mature SC.</p> <p>A detailed study of the cellular and subcellular distribution of AHNAK during development of the rat sciatic nerve was performed on the light and electron microscope levels. During the first post-natal month, AHNAK distribution shifts from adaxonal compartments to abaxonal and outer-mesaxonal SC membranes in contact with basement membrane, and ultimately delineates the so-called “Cajal bands”, exhibiting a staining pattern complementary to that of periaxin. Highly expressed in non-confluent cultured primary SC seeded on laminin, AHNAK is downregulated in confluent cells, mainly concentrated around the nucleus. Furthermore, we noted that the SC laminin receptor beta-dystroglycan exhibits a similar distribution pattern as AHNAK on both confluent and non-confluent cultured SC. AHNAK silencing in cultured SC via siRNA transfection was found to affect the morphology and adhesive properties of SC. This is likely related to our observation that <i>ahnak</i>-siRNA transfection leads to a reduction of beta-dystroglycan expression levels, and dislocation of the receptor from the plasma membrane. Taken together, these results strongly suggest a role of AHNAK in SC interaction with laminin, an important component of the basement membrane surrounding myelinating SC.</p> <p>Loss or mutation of basal lamina components (e.g. laminin-2), or of laminin receptors (integrins and dystroglycan), is known to cause severe demyelinating pathologies affecting the sensori-motor system. Elucidating the role of AHNAK in SC may thus contribute to our understanding of SC differentiation, myelin formation, and myelin maintenance.</p>

PW23-290

**MOLECULAR STUDY OF AUTOSOMAL RECESSIVE HEREDITARY MOTOR AND SENSORY NEUROPATHIES IN A PANEL OF 150 ALGERIAN FAMILIES**

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<p>To contact the author:: tarikhamadouche@hotmail.com.</p>	<p>Hereditary motor and sensory neuropathies (HMSN), commonly referred to as Charcot-Marie-Tooth disease (CMT), are among the most common inherited neurological diseases, with an overall prevalence of about 1-4/10,000. While all modes of inheritance have been reported, clinical, anatomopathological and particularly genetic heterogeneity have also already been underlined for this disease (more than 50 loci and 30 genes identified to date).</p> <p>The aim of this work was to investigate Algerian families for whom a clinical diagnosis of hereditary motor and sensory neuropathy has been suspected. Considering the high rate of consanguinity in Algeria, we focussed our analysis on families affected with autosomal recessive forms of the disease.</p> <p>The use of the strategy of homozygosity mapping, as well as a set of molecular tools, allowed us to explore a panel of 150 families and to characterize for some of them the molecular defect responsible of the observed phenotype, as well as the identification of a new locus whose gene has been further characterized.</p> <p>Thus, we could establish a precise molecular diagnosis for about 1/3 of the families, by characterizing mutations in several genes, <i>MTMR2</i> (CMT4B1), <i>GDAP1</i> (CMT4A), <i>PRX</i> (CMT4F), <i>SH3TC2</i> (CMT4C), <i>GAN</i> (GAN) and <i>LMNA</i> (CMT2B1), whereas one new locus/gene could be identified (CMT4H/<i>FGD4</i>). Although molecular investigations are in progress for the remaining families, our results greatly reinforce the genetic heterogeneity already reported for CMT disease, and suggest that many other loci/genes have to be discovered.</p> <p>This study allowed us to estimate the relative frequency of autosomal recessive forms of CMT disease in our panel, to outline a certain geographical distribution for some CMT subtypes and to delineate the occurrence of recurrent mutations or founder effects. These observations are all important informations that will guide us to establish an efficient approach for diagnosis of autosomal recessive types of CMT disease in Algeria.</p>
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<p>PW23-291</p>	<p><b><u>MECHANISM OF EPISODIC ATAXIA TYPE 2 AND CORRECTION BY STOP-CODON READ-THROUGH STRATEGY</u></b>  MEZGHRANI A<sup>1</sup>, BARBARA G<sup>1</sup>, MONTEIL A<sup>1</sup>, LORY P<sup>1</sup>, NARGEOT J<sup>1</sup>  (1) Institut de Génétique Fonctionnelle, MONTPELLIER, FRANCE.</p>
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Aminoglycosides compounds can read-through to premature termination codon mutations and appear an interesting therapeutic approach for some genetic diseases. Indeed, clinical studies have been initiated for cystic fibrosis and muscular dystrophy diseases. The channelopathy episodic ataxia type-2 (EA2) is an autosomal dominant disorder related to mutations in the pore-forming  $Ca_v2.1$  subunit of P/Q type calcium channels. In addition, P/Q type channels are also the major actors of neurotransmitter release at the neuromuscular junctions. Most of the EA2 mutations are nonsense and lead to the expression of truncated channels that can abolish channel activity via a dominant-negative mechanism (Mezghrani et al, 2008). We have use the EA2 model as a prototype of neuronal and neuromuscular disease in order to evaluate the aminoglycoside-induced read-through approach. Indeed, the EA2 mutant (R1279X) exerts a specific dominant-negative effect on the  $Ca_v2.1$  channel. Using this mutant, we used cytometry-based assay and patch clamp electrophysiology to compare different aminoglycoside compounds for their efficacy to restore functional calcium channels. These data may open new perspectives for future therapeutic strategies for neuronal and neuromuscular diseases.

Mezghrani A, Monteil A, Watschinger K, Sinnegger-Brauns MJ, Barrère C, Bourinet E, Nargeot J, Striessnig J and Lory P. A destructive interaction mechanism accounts for dominant-negative effects of misfolded mutants of voltage-gated  $Ca^{2+}$  channels. *J Neurosci*. 2008, in press.

PW23-292	<p><b>CA-BINDING PROTEINS IN THE SEARCH FOR NEW MODELS OF GLYCINE RECEPTOR POTENTIATION IN HYPEREKPLEXIA</b></p> <p>MUKHTAROV M<sup>1</sup>, BULDAKOVA S<sup>2</sup>, BREGESTOVSKI P<sup>3</sup></p> <p>(1) Institut de Neurobiologie de la Méditerranée (INMED), INSERM U901, Marseille, FRANCE. (2) Institut de Neurobiologie de la Méditerranée (INMED), INSERM U901, Marseille, FRANCE. (3) Institut de Neurobiologie de la Méditerranée (INMED), INSERM U901, Marseille, FRANCE.</p>
To contact the author:: pbrages@inmed.univ-mrs.fr.	<p>Hyperekplexia is a genetic neurological human disease, which is accompanied by muscular hypertonia, hyperreflexia, and hypokinesia. Hyperekplexia originates from a dysfunction of glycine receptor (GlyR) channels, which mediate fast inhibitory synaptic transmission in spinal cord, brainstem, retina and other areas of the nervous system of vertebrates. Several point mutations of the GlyR gene result in decreased activation of GlyR channels, leading to a reduction of inhibitory drive through glycinergic synapses and, consequently, to the development of motor disorders.</p> <p>Previously our team discovered a new mechanism for GlyR modulation: potentiation by intracellular calcium (Ca<sup>2+</sup>) involving a Ca<sup>2+</sup>-binding protein (Fucile et al., 2000). This mechanism could be exploited to increase activity of GlyR channels in glycinergic synapses in normal and pathological conditions.</p> <p>Here we analyzed the mechanisms of Ca<sup>2+</sup>-induced potentiation in neurons and in heterologous systems. We find out that neuronal Ca<sup>2+</sup>-binding protein (NECAB) is involved in Ca<sup>2+</sup>-dependent regulation of GlyRs. Overexpression of NECAB caused: (i) a decrease the apparent affinity to glycine and shift to the right of “dose-response” curve of glycine-induced currents in CHO, HEK-293 cells and in spinal neurons; (ii) a shortening of the decay time of glycinergic currents in spinal neurons.</p> <p>These observations suggest that the development of peptides mimicking the GlyR sequence responsible for binding with NECAB might be important for modulation inhibitory drive through glycinergic synapses. Such peptides, interacting with NECAB, should prevent its binding to the GlyR, thus leading to a functional up-regulation of GlyR channels. Such an approach is a promising avenue to develop pharmacological compounds upregulating GlyRs function in hyperekplexia models.</p> <p>Fucile S., De Saint Jan D., de Carvalho L.P., Bregestovski P., 2000. Fast potentiation of glycine receptor channels by intracellular calcium in neurons and transfected cells. <i>Neuron</i> 28: 571-583.</p> <p><b>ACKNOWLEDGEMENTS</b> This work was supported by French Association against Myopathies (AFM) for M.Mukhtarov.</p>