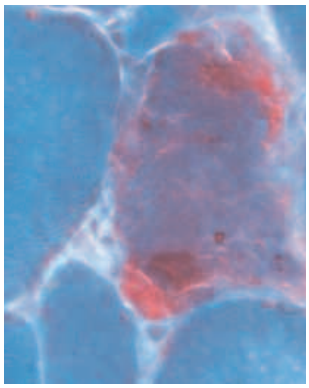
MRC Medical Research Council

Centre for Neuromuscular Diseases

Muscular Dystrophy Campaign






# Inaugural International Neuromuscular Conference

## 1-2 February 2008

UCL Institute of Child Health
30 Guilford Street
London WC1N 1EH

## Contents

## Welcome to the inaugural scientific meeting of the new MRC Centre for translational research in neuromuscular diseases

Dear Colleagues,

I am delighted to welcome you to this inaugural scientific meeting of the first MRC funded centre for translational research in neuromuscular diseases. This new centre aims to bring together clinicians, scientists, patient organizations and patients in order to advance UK translational research in neuromuscular disease. This is a particularly exciting time in the field as a range of basic science discoveries are revealing an increasing number of therapeutic targets. The centre aims to work with all its partners to support the development of a trials culture for patients with neuromuscular diseases. We will work hard to form reciprocal research and clinical links with as many other UK neuromuscular groups as possible.

The newly formed MRC Centre is a joint partnership between the UCL Institute of Neurology, UCL Institute of Child Health, and the University of Newcastle-upon-Tyne. The Centre is closely linked to its partner NHS organizations, University College London Hospitals NHS Foundation Trust, Great Ormond Street Hospital for Children and Newcastle Upon Tyne Hospitals NHS Foundation Trust.

Over the next two days a programme of international speakers will deliver state-of-the art lectures on the scientific foundations of translational research and on the tools required to deliver translational research. There is also an important session which aims to explore how the MRC Centre can contribute to developing UK patient/expert clinician networks. It is these networks that are critical to the delivery of effective translational research and clinical trials. We have received over 50 high quality abstracts and there will be dedicated poster sessions each day.

I would like to thank the joint MRC-MDC planning group for all their hard work organizing this meeting. I am delighted that there has been such an enormous interest in this meeting from throughout the UK.

I sincerely hope that you have a stimulating and entertaining two days in London!

**Professor Michael G Hanna**
Director, MRC Centre for Neuromuscular Diseases
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## Welcome from Mr Philip Butcher

Welcome to the Inaugural International Neuromuscular Conference held in partnership between the new MRC centre and the Muscular Dystrophy Campaign.

The MRC Centre is a scientific translational research partnership between the UCL Institutes of Neurology and Child Health and the University of Newcastle upon Tyne. Not only is such a partnership timely, given the rate of progress shown by researchers working on NMDs, but has as its main goal to accelerate this work by establishing a firm base for trial development.

There are few things closer to the heart of the Muscular Dystrophy Campaign, the other charities and patient groups working together to fight NMD and all the families throughout the country living with these conditions than bringing treatments closer. It is for this reason that all the MDC's supporters and families are excited to support the MRC Centre, support this conference and wish it every success.

One of the rate determining steps to treatment delivery is access to patient cohorts in a planned and structured environment. The model for this is a multi-disciplinary centre or network of excellence with concentrated expertise and critical mass across all NMDs. What we know though is that there is a significant shortfall in centres and networks of this kind in the UK, somewhere in the region of 60% of what is required to serve the relevant patient population.

As well as providing direct financial and other support to existing Centres and Networks the Muscular Dystrophy Campaign is actively working with the NHS and Department of Health to grow their number to meet the known shortfall. A major benefit arising from this will be the acceleration of translational research.

We are very pleased to be jointly hosting this meeting with the MRC Centre for Neuromuscular Diseases and hope that the delegates will use this as a forum to discuss ideas and collaborations, helping to expedite a smooth and speedy transition of promising technology from the "bench" to the "bedside".

**Mr Philip Butcher**
Chief Executive, Muscular Dystrophy Campaign
p.butcher@muscular-dystrophy.org

# The new MRC Centre for translational research in neuromuscular diseases
# A London- Newcastle partnership to form UK-wide translational research networks

**About the Centre**

Genetic and acquired neuromuscular diseases represent a major cause of mortality and morbidity in children and adults. In the UK there is a large gap between major science discoveries and patient benefit in these important disorders. This gap is larger in the UK than in other countries such as Germany, France and the USA who have already moved forward with translational research initiatives. The new MRC Centre aims to reduce this gap by establishing a multidisciplinary translational research activity in these disabling diseases.

This is a joint Centre between the UCL Institute of Neurology and the UCL Institute of Child Health, London and the University of Newcastle. The Centre will build on long established UCL-Newcastle research and clinical links. The centre will form reciprocal clinical and research links with other neuromuscular research groups and patient organizations throughout the UK. The Centre will work with the very large adult and paediatric neuromuscular disease patient populations cared for at the co-located hospitals: Great Ormond Street NHS Trust, the National Hospital for Neurology and Neurosurgery-Queen Square, UCLH NHS Foundation Trust and Newcastle Upon Tyne Hospitals NHS Foundation Trust.

Our mission is to translate basic science findings into clinical trials and new treatments for children and adults with disabling neuromuscular diseases. Current world class science programmes in London and Newcastle attracting in excess of £20m of grant income will underpin the activities of the Centre. The Centre will develop new cross cutting collaborations and will capitalize on the recruitment of world class senior academic personnel to UCL and to the University of Newcastle. We have identified five key areas which we consider to be current obstacles to effective translation of basic science findings into patient benefit. These are; clinical trials support, availability of patient tissues and cells, assessing animal models, applying MRI to humans and animals and developing capacity for the future. Over the next five years the Centre will specifically address each of these obstacles.

- We will facilitate clinical trials in neuromuscular disease in the UK by forming a single clinical trials support activity drawing on and combining the expertise in London and Newcastle. We will take advantage of the geography by forming north and south neuromuscular clinical trials centres. We will work together to facilitate clinical trial design, to develop biostatistical support, to develop clinical trial coordination, and to establish patient registries and clinician networks. We will take advantage of well established, government funded, collaborative specialist neuromuscular diagnostic services which already exist between London and Newcastle (NCG services). In addition, we aim to develop a range of specific clinical assessment tools and outcome measures.
- A shortage of human cell lines and neuromuscular tissues currently hinders basic science efforts and in vitro testing of potential therapies. We will establish a unique UK biobank of human neuromuscular patient tissues and cells.
- Assessing the validity of animal models of neuromuscular disease and correlating phenotypes with human disease remains an important problem. We will link clinical and basic scientists thereby establishing a network and resource for elucidating the validity of mouse models.
- We believe that the application of new MRI techniques has the potential to revolutionize the assessment and monitoring of neuromuscular disease in both animal models and patients. We will take advantage of major new MRI facilities in London and Newcastle to establish cutting edge MRI of nerve and muscle disease in animals and humans.
- We recognize the critical importance of training the basic and clinical neuromuscular scientists of the future. The Centre has developed a brand new four year neuromuscular disease PhD programme. We will deliver exciting translational research environments to attract and train a new generation of basic and clinical neuromuscular scientists to build future capacity in the UK.

By developing these five core areas the Centre will promote translational research and add value to basic science neuromuscular research themes currently active in London, Newcastle and other centres.

## About the Muscular Dystrophy Campaign

The Muscular Dystrophy Campaign is the only national charity focusing on all muscular dystrophies and allied disorders. It has pioneered the search for treatments and cures for nearly 50 years and provides practical, medical and emotional support to people affected by the condition. Our mission is to meet the needs of people with neuromuscular conditions in the UK, their carers and their families. We will do this by searching for treatments and cures for all the conditions, by providing excellent support services and by enabling and empowering the people we exist to help.

The Muscular Dystrophy Campaign has, since it was founded in 1959, supported scientists in the UK working on muscle disease in their effort to find the underlying molecular basis of muscular dystrophies and allied disorders. In recent years, the focus of this research has begun to shift towards the search for treatments for these conditions.

Translational research involves a two way interaction between the scientists and the clinicians. The basic bench science is important for understanding underlying causes of disease, something that can provide a plethora of potential drug or gene therapy targets. Equally the observations that the clinicians make at the bedside can provide a wealth of new information about a condition focussing the search for the scientist. There are however, many barriers in the meaningful progression of data and observations from the lab to something that will ultimately benefit the patient.

The Muscular Dystrophy Campaign aims to ease this transition by providing support to both scientists and clinicians. We fund basic research through to pre-clinical research and, where possible, clinical trials. As well as monetary help we aim to provide a platform where clinicians and scientists can meet and discuss ideas.

One of the charity's strategic aims is to fast-track promising treatment approaches when they are close to clinical trial and to ensure a rapid transition from bench to bedside. A major focus of the last year has been to support and encourage initiatives to promote translational research in order to help remove some of the barriers faced by scientists and clinicians. The charity currently supports research across a range of disciplines from the basic science through to research looking at treatment approaches for a number of conditions.

In order to give you an overview of the science that the charity supports, we have invited our current grantees to present their work during the poster sessions. We hope that these sessions will be of interest to you and provide you with an update of the recent research advances that the scientists and clinical researchers have made.

## Inaugural International Neuromuscular Conference Programme

## London-Newcastle MRC centre for translational research in neuromuscular disease

The new MRC centre is a scientific translational research partnership between the UCL Institutes of Neurology and Child Health and the University of Newcastle upon Tyne

**Friday 1 February 2008**

**9.00** Registration

**The stakeholders in translational research**
**Chaired by Professor Mike Hanna and Mr Philip Butcher**

**10.00** Professor Mike Hanna
Welcome, introduction and overview of the centre and UK networks

Professor Malcolm Grant
Welcome from UCL's President and Provost

**10.15** Professor Edward Byrne
Translational research across UCL

**10.30** Professor Kate Bushby
Newcastle partner centre

**10.45** Mr Philip Butcher
Neuromuscular patient groups

Dr Adrian Pollitt OBE
National Specialised Commissioning

**11.00** Professor Sir Leszek Borysiewicz
Translational research in the UK

**11.20** Coffee and Posters

**12.00** The Victor Dubowitz Lecture

Professor Robert Griggs
Translational research in neuromuscular disease - Basic science to clinical trials & new treatments

**12.45** Lunch and Posters

**Basic science foundations of translational research**
**Chaired by Professor Francesco Muntoni and Dr Mary Reilly**

**1.45** Professor Eric Hoffman
Genome-enabled progress in muscular dystrophy pathophysiology and therapeutics

**2.15** Professor Thomas Voit
Spinal muscular atrophy

**2.45** Professor Mike Shy
Myelin protein zero in myelination and disease

**3.15** Coffee and Posters

**Chaired by Professors Mike Hanna and Dimitri Kullmann**

**4.15** Professor Stephen Waxman
Aberrant excitability: the molecular pathology of neuromuscular disease

**4.45** Professor Louis Ptácek
Molecular characterization of episodic neurological disease

**5.15** The First Morgan-Hughes-Thomas Lecture

Professor Salvatore DiMauro
Human mitochondrial disease and translational research

**6.00** Posters and Drinks Reception

**7.45** Dinner at The Honourable Society of Lincoln's Inn

## Saturday 2 February 2008

**Basic science foundations of Translational research**
**Chaired by Professors Douglass Turnbull and Volker Straub and Dr Mary Reilly**

**9.00** Professor Vincent Timmerman
Are ubiquitously expressed genes good candidates for CMT neuropathies?

**9.30** Professor Marinos Dalakas
Interrelationship between degenerative and inflammatory pathways in sIBM muscle and therapeutic implications

**10.00** Coffee and Posters

**Translational Research - Europe and USA**
**Chaired by Professors Mike Hanna and Francesco Muntoni**

**10.45** Professor John Porter
Experiments in public support of translational research in neuromuscular disease - an NIH (USA) perspective

**11.15** Professor Serge Braun
European translational research

**11.45** Professor Kate Bushby
TREAT-NMD - an EU network of excellence to accelerate treatments for rare inherited neuromuscular diseases

**12.05** Lunch and Posters

**The networks to deliver translational research**
**Chaired by Mr Robert Meadowcroft and Professor Mike Hanna**

**1.05** Mr Nick Catlin, Action Duchenne
**1.15** Mrs Kate Parkin, Duchenne Family Support Group
**1.25** Mrs Sheila Hawkins, FSH Support Group
**1.35** Dr Peter Streng, EAMDA
**1.45** Dr Douglas Wilcox, Scottish Muscle Network
**1.55** Mrs Elaine Scott, NorthStar Clinical Network
**2.05** Professor Mike Hanna, British Myology Society
Dr Mary Reilly, British Peripheral Nerve Society

Panel Members:
Professors Kate Bushby, Francesco Muntoni, Patrick Chinnery, Volker Straub and Dr Mary Reilly

**2.20** Posters and coffee

**The tools of translational research**
**Chaired by Professors Martin Koltzenburg, Hanns Lochmüller and Tarek Yousry**

**3.00** Professor Francesco Muntoni
Biobanking tissue for translational research in the UK

**3.30** Professor Martin Bendszus
Novel MR techniques in neuromuscular disease

**4.00** Professor Richard Hughes
The Cochrane Collaboration Contribution

**4.30** Professor Mike Hanna and Professor Martin Bobrow
Closing comments and poster prize giving

# Speaker Abstracts

**The Victor Dubowitz Lecture**

**Translating basic discoveries into standard treatment for neuromuscular disease**

Robert C. Griggs, M.D.
*Professor and Chair of Neurology, University of Rochester School of Medicine and Dentistry*

Molecular discoveries have identified the cause of most neuromuscular diseases and provided almost unlimited opportunities for developing treatment strategies for these diseases. The challenge now is to overcome the obstacles to translating these opportunities into therapies that are evidence-based, available and affordable world wide. There are currently few approved and no standard treatments. Most neuromuscular diseases are rare and the molecular heterogeneity of "private" mutations further subdivides diseases by phenotypic heterogeneity. Establishing the benefit of a novel treatment will require: large networks of specialist physicians; careful characterization of the natural history of each untreated disease; the development of outcome measures that demonstrate treatment benefit and result in patient satisfaction and improve quality of life. The obstruction to accomplishing these goals loom large: The need for trial designs appropriate for rare diseases; the need for harmonization of ethics/human subjects review standards; harmonization of the drug regulatory approval procedure; the competition for research funding; the recruitment, support and training of the next generation of clinical scientists. There are encouraging signs and reasons for optimism. The harmonization process has begun. The regulatory approval process is changing. Industry is investing in neuromuscular diseases. Expensive treatments are being paid for. Effective patient advocacy groups are getting attention and private philanthropy is expanding support for research in even the rarest diseases. However, ultimate success requires that each of the steps involved in translating science to benefit patients be kept in view and given attention.

**Genome-enabled progress in muscular dystrophy pathophysiology and therapeutics**

Eric P Hoffman, Akanchha Kesari, Zuyi Wang, Toshifumi Yokota, Kanneboyina Nagaraju, Shin'ichi Takeda.
*Research Center for Genetic Medicine, Children's National Medical Center, Washington DC*
*National Center for Neuroscience and Psychiatry (NCNP), Kodaira, Japan*

It is now 20 years since the identification of the dystrophin gene and protein, and a complete understanding of the biochemical defect causing the most common type of muscle disease, Duchenne muscular dystrophy. Sporadically occurring animal models including mice, dogs and cats were quickly identified, and studies of molecular pathophysiology downstream of dystrophin loss at the membrane, and experimental therapeutics proliferated quickly. Here, we present data from a series of unpublished studies on molecular pathophysiology and therapeutics. The results of integrated DNA, cDNA and protein studies in 75 Becker muscular dystrophy samples showed high exception to the reading frame rule (30%), in part due to removal of additional exons in the mRNA. This finding suggests that the study of genomic deletions alone may not be adequate to predict the consequences on mRNA and protein. In this study, we also describe a male somatic mosaic for a nonsense mutation. This young man presented with cardiac failure following shoveling of snow. After successful cardiac transplant he showed persistent CK elevations, leading to our eventual diagnosis of a somatic mosaic. In the second set of studies, we identify compensatory pathways invoked by muscle in response to dysferlin deficiency; this leads to a model of interaction between these compensatory exocytotic vesicle trafficking pathways and the immune system, explaining the sub-acute and inflammatory onset experienced by some LGMD2B patients. We then turn to explaining the beneficial effect of glucocorticoids on Duchenne dystrophy patients; using a data integration approach, we present a model where daily glucocorticoids serve to re-synchronize muscle regeneration in dystrophic muscle. Finally, we present therapeutic studies of systemically administered morpholinos (exon skipping) in the large animal dog model of Duchenne muscular dystrophy. In studies of three dogs, treated up to 6 months, intravenous delivery of grams of a cocktail of three morpholinos stabilized or improved multiple measures of muscle function.

**Myelin protein zero in myelination and disease**

Michael E. Shy, MD
*Professor of Neurology and Genetics, Wayne State University, USA*

Myelin protein zero (MPZ), the major structural protein in PNS myelin, is a homotypic adhesion molecule necessay for normal myelin compaction. MPZ is the simplest member of the imunoglobulin supergene family and consists of three structural domains: a single 124 amino acid immuno-globulin-like extracellular domain; a 26 amino acid transmembrane domain; and a 69 amino acid intracellular domain. Crystallographic analysis of the MPZ extracellular domain suggests that the protein interacts within the plane of the membrane to form a lattice of homotetramers, each of which then interacts with similar structures on the opposing myelin loops to mediate myelin compaction. The cytoplasmic domain of MPZ is also necessary for its adhesive function, since deletion of the carboxy terminal 28 amino acids, including a putative PKC$\alpha$ target site, abolishes MPZ-mediated adhesion *in vitro*. Recent data shows that this portion of the protein interacts directly with both RACK1 and PKC$\alpha$ to regulate phosphorylation and homotypic adhesion, demonstrating that MPZ, like other Ig superfamily members, participates in a signal transduction cascade.

More than 120 different *MPZ* mutations have been identified in humans that cause the hereditary demyelinating neuropathy, Charcot Marie Tooth disease type 1B (CMT1B) Interestingly, most patients with neuropathy caused by *MPZ* mutations can be separated into two distinct groups: dysmyelinating neuropathies in which disease onset occurs in infancy, the clinical disability is relatively severe, and nerve conduction velocities (NCV) are very slow < 15 m/s); and a second in which disease onset occurs in adulthood, usually after age 40, the clinical disability is relatively mild, and nerve conduction velocities are essentially normal. Morphological studies of these early and late onset forms of CMT1B have demonstrated distinct molecular and pathological abnormalities, confirmed by studies in tissue culture and in "knockin mice". Analysis of these neuropathies has provided essential information concerning the role of MPZ in myelination and in the pathogenesis as well as treatment strategies for "gain of function" inherited neuropathies like CMT1B.

**Aberrant excitability: the molecular pathology of neuromuscular disease**

Stephen G Waxman, M.D., Ph.D.
Bridget Marie Flaherty Professor of Neurology, Neurobiology and Pharmacology
*Chairman, Dept. of Neurology, Yale University School of Medicine*
*Director, Center for Neuroscience & Regeneration Research, VA Medical Center, West Haven, CT*

Voltage-gated sodium channels are widely expressed within neurons and play pivotal roles in neuronal signaling. Less has been known about the roles of sodium channels in disorders of the spinal cord and brain. Neurophysiology classically referred to "the" sodium channel as if it were a singular entity. Recent research has taught us that sodium channels are more complex than previously appreciated, and has shown us that sodium channels play multiple roles in neuronal pathophysiology.

In this lecture I will review the following advances: 1) we now understand that ten different genes encode ten different isoforms of sodium channels, with different kinetic and voltage-dependent properties. The presence of different ensembles of sodium channel isoforms in different types of neurons endows them with different firing properties. 2) sodium channel expression is not static. It is plastic, in both the normal nervous system and, to a greater degree, in the injured and diseased nervous system. 3) some changes in expression of sodium channels are maladaptive, an example being neuropathic pain, where dysregulated expression of sodium channels can lead to abnormal high-frequency firing of neurons along the pain-signaling pathway in the absence of external painful stimuli. 4) we now understand, for the first time, a human hereditary pain disorder (erythromelalgia, the "man on fire syndrome") which is due to a gain-of-function mutation in a sodium channel (Nav1.7) that makes nociceptive DRG neurons hyperexcitable, providing a model in humans of chronic pain due to sodium channel dysfunction. 5) The pivotal role of sodium channels as generators of hyperexcitability underlying chronic pain, and the selective expression of certain sodium channel isoforms in nociceptors, makes them attractive therapeutic targets.

**References:**

Waxman, S.G., Channel, neuronal, and clinical function in sodium channelopathies: From genotype to phenotype. Nature Neuroscience, 10:405-410, 2007.
Waxman. S.G., Hains, B.C. Fire and phantoms after spinal cord injury: sodium channels and central pain. Trends in Neurosciences 29: 207-215. 2006
Waxman, S.G., Dib-Hajj, S.D. Erythermalgia: molecular basis for an inherited pain syndrome,
Trends in Molecular Medicine, 11 (12): 555-562, 2005

**Molecular characterization of episodic neurological disease**

Louis Ptácek
Investigator, Howard Hughes Medical Institute,
*John C Coleman Distinguished Professorship in Neurodegenerative Diseases, Department of Neurology, Mission Bay Campus, Rock Hall, Room 548F*

Episodic neurological phenotypes are a very interesting and important group of diseases affecting humans. These include disorders of skeletal and cardiac muscle, peripheral nerve, and brain. They range from episodic weakness syndromes to paroxysmal movement disorders that are quite rare. We have shown that ion channel gene mutations are responsible for many of these disorders but have also cloned novel genes causing epilepsy and paroxysmal dyskinesias that do not encode ion channels. One of these genes encodes a protein that we predict to function in stress response; this raises the possibility that proteins critical for homeostatic responses to altered membrane excitability might be sights for genetic variants affecting risk for episodic disorders. More common episodic phenomena include cardiac arrhythmias, epilepsy syndromes, and headache. In rare Mendelian disorders, single gene mutations are sufficient to cause dramatic phenotypes. Knowledge gained from molecular characterization of rare genetic disorders is informing studies of the genetically and clinically more complex diseases. Molecular characterization of all of these disorders is shedding light on pathophysiology and will ultimately lead to better diagnosis and treatment of patients.

**The First Morgan-Hughes-Thomas Lecture**

**Human mitochondrial disease and translational research**

Salvatore DiMauro, MD
*Department of Neurology, Columbia University Medical Center, New York, NY, USA*

Exactly 20 years ago, a seminal paper by Ian Holt, John Morgan–Hughes and Anita Harding opened the molecular era of research in human mitochondrial disease. The discovery of pathogenic mutations in mitochondrial DNA (mtDNA) had a translational ripple effect that is still expanding. Intensive sequencing of mtDNA revealed many more pathogenic mutations than anybody expected and revolutionized the way we approach inherited diseases (mitochondrial genetics) and the way we reason diagnostically (mitochondrial medicine).
A second translational ripple was the realization that mtDNA is the "slave" of nuclear DNA (nDNA) and led to the discovery of an interesting subgroup of Mendelian mitochondrial diseases due to garbled "dialogue" between the two genomes. These disorders come in at least five flavors: (i) defects of respiratory chain (RC) assembly proteins; (ii) defects of mtDNA integrity; (iii) defects of mtDNA replication; (iv) defects of mtDNA translation; (v) defects of the inner mitochondrial membrane lipid composition.
Although the first two ripples both involve RC function directly or indirectly, mitochondria are much more than the "powerhouses of the cell" described in textbooks, as they also allocate energy to different cell compartments through their until recently unsuspectedly lively dynamic behavior; they contribute to calcium homeostasis; and they preside over at least one form of controlled cell death (apoptosis).
Thus, a third translational ripple has recently taken center stage, involving disorders due to defective mitochondrial motility, fusion, or fission. Not surprisingly, these seem to affect predominantly the central and peripheral nervous system and may play a role in the pathogeneses of late-onset neurodegenerative diseases.
The ultimate translational ripple is – of course – therapy. Sadly, this is still woefully inadequate, although ingenious therapeutic strategies are being developed, at least in the laboratory. But this is another story deserving a separate lecture.

**Are ubiquitously expressed genes good candidates for CMT neuropathies?**

Vincent Timmerman, PhD
*Molecular Genetics Department, VIB, University of Antwerp, Belgium*

The most common inherited peripheral neuropathy is Charcot-Marie-Tooth (CMT) disease, which is characterized by progressive weakness and atrophy of foot and hand muscles, with some patients becoming severely affected at a young age. Positional cloning has already identified more than 40 loci and 35 genes for dominant, recessive or X-linked forms (**www.molgen.ua.ac.be/CMTMutations**). Identification of these genes is a first step towards a better understanding of the fundamental biological processes operating in myelination, axon-Schwann cell interactions, structure of the axonal cytoskeleton and axonal transport. We have characterized several large families that represent novel genetic entities, and described mutations in distinct genes leading to demyelinating and/or axonal forms of CMT. A number of these genes (e.g. PMP22, MPZ, GJB1) are expressed in Schwann cells, while others are expressed in the neuron (e.g. NEFL). Interestingly some genes are ubiquitously expressed, such as the small heat shock proteins HSPB1 and HSPB8 in which mutations result in distal motor neuropathy or axonal CMT. Mutations in the small heat shock proteins cause protein aggregation, alteration of the neurofilament assembly and cell death. Also mutations in the aminoacyl-tRNA-synthetases, GARS and YARS, which are essential enzymes in the biosynthesis of cellular proteins, have been associated with distinct types of CMT neuropathies. Why mutations in these essential genes cause specifically length-dependent peripheral neuropathy remains enigmatic. We will try to provide some answers to these molecular pathomechanisms in relation to CMT and related neuropathies.

**Interrelationship between degenerative and inflammatory pathways in sIBM muscle and therapeutic implications**

Marinos C. Dalakas M.D.
*Director of the Division of Neuromuscular Diseases in the Department of Neurology and Professor of Neurology at Jefferson Medical College of Thomas Jefferson University, Philadelphia*

In sporadic Inclusion Body Myositis (sIBM), the most prevalent acquired myopathy above the age of 50, chronic inflammatory features co-exist with degeneration. The inflammation is characterized by upregulation of proinflammatory cytokines and chemokines and by clonal expansion of CD8+ T-cells that invade and damage MHC-1-overxpressing muscle fibers. The degeneration is highlighted by the accumulation of β-amyloid derived from amyloid precursor protein (APP), and by cell-stress or degeneration-associated molecules, such as αB-crystallin, tau and ubiquitin, which can impair cellular function and trigger, in vitro, intracellular protein aggregation. Because a similar stressor effect on muscle fibers is also caused by chronic overexpression of MHC-1, the interrelationship between inflammatory and degeneration-associated molecules was explored in the patients' muscles and cultured myotubes. The clinical significance of these molecules was further examined on the repeated muscle biopsies from sIBM patients treated with CAMPATH, a T cell-depleting monoclonal antibody.
The mRNA-expression of CXCL-9, CCL-3, CCL-4, IFN-γ, TNF-α and IL-1β was upregulated in sIBM compared to control muscles. Although the degeneration-associated markers were overexpressed in all myopathic controls, only in sIBM the mRNA of APP significantly correlated with endomysial inflammation and with the levels of chemokines and IFN-γ. Cytokines and stressor

proteins, such as IL-1β, iNOS and αB-crystallin, co-localized with β-amyloid and MHC-1. In cultured myotubes, IFN-γ, TNF-α and IL-1β enhanced the production of cytokines, augmented the expression of APP, _-amyloid and iNOS, and led to intracellular protein aggregation and cell death. Collectively, the data suggest that inflammatory and degenerative mechanisms act in concert to exert myocytoxicity in sIBM. The observations may help design treatment strategies to suppress muscle degeneration by targeting the chronic inflammatory response. This conclusion is further supported by the clinicopathologic observations from the CAMPATH trial.

**Experiments in public support of translational research in neuromuscular disease - An NIH (USA) perspective.**
John D. Porter.
*Neurogenetics Cluster and Technology Development Program, National Institute of Neurological Disorders and Stroke (NINDS), National Institutes of Health (NIH), Bethesda, Maryland, USA.*

Progress in neuromuscular disease mechanisms has identified novel targets and created unparalleled opportunities for therapy development. Yet, gaps exist between mechanistic studies supported by public funds and iterative, therapy development activities of biotechnology and pharmaceutical companies. For rare disorders, including spinal muscular atrophy (SMA), inherited neuropathies, channelopathies, mitochondrial and inflammatory myopathies, and muscula dystrophies, it falls to federal agencies and patient organizations to bridge the gap where industry incentives for early-stage development efforts may be weak or non-existent.
Success in translational research will require changes in investigator and funder mindsets, partnerships, and funding paradigms. NIH has initiated workshops and funding paradigms that represent, in part, 'experiments' in process change and in moving therapeutic candidates toward clinical trials. These experiments in discovery and development include: (a) NINDS SMA Project, which adopted pharmaceutical industry methodology and is now licensing its first development candidate, (b) NIH Wellstone Muscular Dystrophy Cooperative Research Centers, supporting a traditional centers of excellence model integrating preclinical research, patient-oriented research, and infrastructure, (c) NINDS Cooperative Program in Translational Research using a milestone-driven model for preclinical testing of therapeutic strategies for neuromuscular disease, and (d) strategic planning and priority setting workshops that address partnering and changes in mindset. Such diversity in the NIH research portfolio is essential, as it is unclear which paradigm(s) will best accelerate the development of novel therapeutics. These programs cannot succeed in isolation. Emergence of therapies for currently untreatable diseases ultimately will require international academic-corporate-government-patient group partnerships to share expertise, resources, and risks in order to reduce the clinical burden of neuromuscular disease.

**European translational research**
Serge Braun
*AFM, France*

For a number of diseases, large networks must be established at a level far beyond regional or national levels. One of the reasons is the necessity to reach critical masses of patients and dedicated researchers. This is particularly true in the neuromuscular field, especially due to the considerable heterogeneity in prevalence, genotypic/phenotypic features as well as in the means and fund raising capacity of the various patient associations.
Very early-on (immediately after the first French Telethon in 1987), AFM became convinced of the need to bring together researchers from different countries on one hand and specialists of different disciplines on the other hand: basic scientists, geneticists, clinicians, and, more recently, professionals of clinical development. This strategy led to major breakthroughs in the field of muscle diseases and new therapeutic means, as illustrated by the explosion of knowledge and publications in myology. With special emphasis on rare neuromuscular diseases, European organisations such as Eurordis and the European NeuroMuscular Center (ENMC) have been catalysers of this model of translational research. It elicited the emergence of new myology teams all over Europe.
Some of those consortia have been funded by the European Commission. Several challenges however remain: socio-economical issues, private-public partnerships, the not yet finalised recognition of Expert Centres, the integration of new countries in the European Union, their patients as well as their too rare specialised clinicians and researchers.

**TREAT-NMD - an EU network of excellence to accelerate treatments for rare inherited neuromuscular diseases**
Kate Bushby and Volker Straub
*Institute of Human Genetics and MRC Centre for translational research in neuromuscular diseases, Newcastle upon Tyne*

An EU network of excellence is an instrument designed to help researchers work together towards a common goal. In the last round of Framework 6 calls, the EU called for applications for a network of excellence to address the challenges faced in implementing cutting edge treatments for rare inherited neuromuscular diseases. Supported by AFM and other patient organisations, a bid led from the University of Newcastle and comprising 21 European partners was successful and TREAT-NMD was launched in January 2007. Sixteen activities within the network address issues of "trial readiness" in neuromuscular diseases, with the initial focus on Duchenne Muscular Dystrophy and Spinal Muscular Atrophy. International registries for these disorders are being established

with collaborations extending to 19 different national registy initiatives. Standards of care for SMA were generated in collaboration with the ICC for SMA and DMD care standards are in preparation, with a major emphasis of the network being in the translation and dissemination of these care standards. An active trial co-ordination centre has identified over 70 possible trial sites, and work to generate and standardise outcome measures across these sites is ongoing. A registry of the different trial regulations across Europe is under development, and dialogue is open with regulatory authorities to provide a greater impetus to trial development in these areas. Standardisation of assessment of animal models is also being developed, along with feasibility studies of production and toxicology studies across Europe.
Industrial interest in the network is high, and input from industry and patient groups is a core part of the network, providing very useful direction to its activities. TREAT-NMD is not an exclusive network, and we are always happy for contact from any interested parties. Our newsletter circulation is over 2,500, and our collaborations extend all over the world. Further information can be found at **www.treat-nmd.eu**

**Biobanking tissue for translational research in the UK**

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The rarity and genetic heterogeneity of neuromuscular disorders (NMD) has hampered the understanding of their pathogenesis and the development of therapies. The lack of access to patient tissue and patient derived primary cell cultures for many basic scientists has also slowed down progress. Recent advances have led to the discovery of the majority of the genes responsible for NMDs and to the elucidation of mechanisms involved in these diseases, which could be targeted by specific interventions. In order to take advantage of the large neuromuscular patient population seen at the MRC Centre in London and Newcastle, and to facilitate the translational research activities of the Centre, we have decided to create a repository of human muscle and non-muscle cells from patients with NMD (biobank). In particular we intend to collect tissues and primary cell cultures from skin; muscle; stem cells; and nerve from patients with genetically determined NMDs. A number of projects already initiated (such as the antisense oligonucleotide approach to induce exon skipping in Duchenne muscular dystrophy) are dependent on the availability of such cell cultures and future projects will use this material to investigate biomarkers predictive of therapeutic response in therapeutic trials and analyse the stratification of response.
The ultimate goal of this biobank is to provide an ethically robust and durable framework to collect unique material and make it available to investigators involved in the MRC Centre, but also facilitate access to collections by UK partners and other EU partners. Similar biobanks already exist in other EU countries, and our initiative will be part of a wider European Network, Eurobiobank, which, together with the EU Network of Excellence TREAT-NMD, will facilitate collaboration and translational research in the UK and Europe.

**Novel MR techniques in neuromuscular disease**

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Currently, the evaluation of peripheral nerve and muscle disorders depends on clinical examination, supplemented by electrophysiological studies. These approaches provide general information on the distribution and classification of nerve lesions—for example, axonal versus demyelinative—but morphological and pathophysiological detail is lacking which would require a nerve or muscle biopsy. In this survey, recent progress in the imaging of peripheral nerve injury and neurogenic muscle disorders by magnetic resonance neurography (MRN) will be reviewed. Axonal nerve injury leads to Wallerian degeneration, resulting in a hyperintense nerve signal on T2-weighted (T2-w) MR images of the distal nerve segment, which recovers on successful regeneration. Concomitant denervation-induced signal alterations in muscles can further aid differentiation between affection of nerve trunks and roots. However, these signal changes are caused by various combinations of nonspecific tissue alterations, and are not related to particular patho-anatomical findings. New experimental MR contrast agents allow visualization of the dynamics of peripheral nerve injury and repair. Further clinical development of these MR contrast agents should allow these functional aspects to be assessed in humans, and aid the differential diagnosis of peripheral nerve disorders.

**The Cochrane Collaboration Contribution**

Richard Hughes MD FRCP FMedSci
Emeritus Professor of Neurology, King's College London

The Cochrane Collaboration aims to provide an accessible source of the best evidence on which to base clinical practice. The Neuromuscular Disease Group, one of 51 which between them cover the whole of Medicine, was founded in 1998 and has now published 60 systematic reviews in the Cochrane Library. The editorial base expenses are funded by the Department of Health, recently supplemented by a contribution from the European Union TREAT-NMD programme. However the Editorial Board and review authors work without remuneration for the greater good. Some governments, including those of England, Wales and Scotland, provide free access to their citizens. The publishers working with the Cochrane Collaboration have

provided free access to the Cochrane Library so that more than half the world's population has one click access to the Cochrane Library on the world wide web. Cochrane reviews have earned a reputation for being carefully researched, unbiased pieces of work. This reputation is based on a methodology which has been refined over the past 15 years. The premise is that in most clinical situations the best evidence comes from randomised controlled trials. The Collaboration has devised techniques for collecting, assessing and summarising the evidence in an unbiased way. There is a requirement to set up a hypothesis and declare the search strategy, types of evidence to be included, methods of data collection, and methods of data synthesis in advance as a peer-reviewed protocol.

Resultant reviews may be packed with trials. Chalk and colleagues reviewed aldose reductase inhibitors for treating diabetic neuropathy, identified 31 trials and concluded that there no statistically significant difference from placebo. Others have relatively few trials. Manzur and colleagues reviewed the few randomised trials of corticosteroids for Duchenne muscular dystrophy and discussed the non-ramdomised evidence in an influential review which they have just updated. Some reviews find no randomised evidence but can propose the trials that are needed and the most appropriate patient groups and outcome measures to consider. Cochrane reviews offer evidence but do not make recommendations. However they are often, and appropriately, used as an important source of evidence on which guidelines are based, such as the UK guidelines for intravenous immunoglobulin in neurological disease.

The Cochrane Collaboration is a Sisyphean but fascinating and enjoyable task because the mountain of evidence is thankfully growing fast so that reviews have to be updated frequently. Those who would like to help are invited to contact us:
**www.kcl.ac.uk/schools/medicine/depts/clinneuro/cochrane/about/articles.html**

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# Poster Abstracts

**3. A new mouse model for 'dystroglycanopathies' associated with mutations in Fukutin Related Protein**

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Mutations in fukutin related protein (FKRP) give rise to a broad range of clinical phenotypes all of which are associated with a reduction in the glycosylation of α-dystroglycan in skeletal muscle. Patients at the severe end of the clinical spectrum have more significant disruption of α-dystroglycan glycosylation and associated structural brain and eye involvement. The defective glycosylation of α-dystroglycan results in loss of binding to various extracellular matrix proteins including laminin, agrin and perlecan in muscle and also neurexin in brain. To investigate the mechanism of disease in FKRP related muscular dystrophies, we generated two lines of mice, the first containing a missense mutation and neomycin cassette, FKRP-Neo$^{Tyr\ 307Asn}$ and the second containing the FKRP-Neo$^{Tyr\ 307Asn}$ mutation alone. This mutation, in the homozygous state has previously been associated with Muscle Eye Brain Disease in the human. Heterozygote FKRP-Neo$^{Tyr\ 307Asn}$n mice are indistinguishable from wild type and are fertile but homozygotes die at or soon after birth. The skeletal muscle of these mice shows a marked reduction in the laminin-binding epitope of a-dystroglycan and a reduction of laminin α2 but displays a mild pathology. In addition these mice display marked structural brain defects and abnormal eye vasculature. By contrast FKRP-Neo$^{Tyr\ 307Asn}$ mice show no discernible phenotype. We were able to demonstrate that the mice homozygous for FKRP-Neo$^{Tyr\ 307Asn}$ had greatly reduced levels of FKRP transcript. These observations demonstrate that FKRP plays a fundamental role in muscle, eye and brain development and that mechanisms that alter the expression levels of this protein represent a mechanism of disease in FKRP related dystroglycanopathies.

**4. A model of human muscle regeneration in vivo to test potential therapies for Duchenne muscular dystrophy**

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At present there is no effective treatment for the lethal muscle wasting disorder Duchenne muscular dystrophy (DMD). Numerous potential therapies have been suggested and evaluated in some of the models available but there has not yet been a systematic comparison of the various therapeutic methods. The aim of this project is to use the available small animal models of DMD to assess a number of potential therapies. The focus will be on cell based therapies and the use of antisense oligonucleotides (AONs) to correct out of frame mutations in the dystrophin gene by exon skipping. So far, primary cultures derived from normal and DMD patient muscle biopsies have been characterised in vitro including fibroblasts, myoblasts, pericytes, synovial membrane mesenchymal cells and AC133+ cells from peripheral blood and skeletal muscle. These have been used as a model to test a number of exon skipping strategies in vitro. In addition these cells have been grafted into mouse models to find the optimal cell type and route of administration for transplanting human cells to maximize muscle regeneration in host mouse muscle. This model tissue will be used to attempt exon skipping in vivo in mosaic human/mouse muscle fibres. We will use AONs with a morpholino backbone or an U7-lentiviral vector to correct the mutation in DMD patient cells for use in an ex vivo autologous cell transfer therapy.

**5. Characterisation of a novel sarcomeric Z-band associated complex**

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The KY protein underlies a form of muscular dystrophy in the mouse but its role in muscle remains elusive. KY interactions previously identified included fragments of filamin C and of a novel protein termed KIP (KY Interacting Protein). Characterisation of the KIP locus exposed that at least three distinctive muscle proteins are encoded here: KIP1, KIP2 and KIP2a. Interaction assays within the yeast two-hybrid system supported that KIP1, KIP2, KY and FLNC are part of a protein complex. Furthermore, immunofluorescence analysis of adult skeletal and heart muscles as well as transductions of cardiomyocytes revealed co-localization of KIP1, KIP2, KY and FLNC at the sarcomeric Z-band. A potential signalling role for this complex is suggested by the fact that endogenous KIP1 also shows nuclear localization and that in the absence of the KY protein, there is a significant reduction of nuclear KIP1 in all adult muscles, a molecular feature particularly exacerbated in dystrophic soleus muscle from ky/ky mice. On the other hand, constitutive RNAi-mediated down-regulation of KIP2 in C2C12 myoblasts does not affect proliferation rates. However, once initiated to differentiate these cells fail to fuse and detach from the plate, suggesting that KIP2 has a crucial role at an early stage of the muscle differentiation pathway.

**6. Deteriorating haemodynamics and myocardial injury after prednisolone therapy in muscular dystrophy associated cardiomyopathy**

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By extrapolation from experience in patients with dystrophinopathy, it is thought that oral corticosteroids have beneficial effects on muscle strength in patients with Limb girdle muscular dystrophy type 2F (LGMD2F), whereas so far the effects on cardiac function are not fully understood. To study the effect of oral corticosteroids on cardiac function, we orally treated 8 week old delta-sarcoglycan deficient mice (Scgd null), a model for LGMD2F, with prednisolone (1.5 mg/kg/day) for 8 weeks. Assessment of in vivo cardiac function at the end of the treatment period was done by pressure-volume loops using a conductance catheter. Scgd null cardiomyopathy is well compensated at baseline with decreased myocardial contractility (preload-recruitable-stroke-work), though increased preload (maximal volume) and decreased afterload (arterial elastance) maintaining a high cardiac output. After prednisolone the increased preload and reduced afterload are no longer present and there is prolonged relaxation, thereby reducing cardiac output. On histology after steroid treatment there was increased myocardial cell damage reflected by higher Evans blue dye uptake (sarcolemmal injury) and increased myocardial fibrosis. In conclusion, prednisolone leads to a decompensation of cardiac haemodynamics associated with sarcolemmal injury and myocardial fibrosis, suggesting that clinical trials must closely evaluate cardiac effects of steroids in these patients.

**7. No evidence for involvement of the feline or bovine spinal muscular atrophy genes in human motor neuron disorders**

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Loss of large motor neurons in the anterior horn of the spinal cord, with little or no involvement of other systems, is the hallmark of human spinal muscular atrophy (SMA). While the majority of cases are an autosomal recessive disease due to homozygous deletion of the Survival Motor Neuron locus on chromosome 5q13, a minority of patients are clinically and genetically heterogeneous, with no known causative gene in many cases. The loss of lower motor neurons is a feature shared by amyotrophic lateral sclerosis (ALS), a disease that in addition affects the upper motor neuron in the cerebral cortex. Cats and cows develop spinal muscular atrophy that is pathologically indistinguishable from the human disorder, as an autosomal recessive trait. The causative genes have recently been identified by positional cloning as LIX1 and FVT1, respectively. To investigate whether LIX1 or FVT1 are contributing to human motor neuron disorders, we screened 96 patients with non-5q SMA and 119 patients with non-SOD1 ALS for mutations in the human orthologues of cat LIX1 and bovine FVT1 using denaturing high-performance liquid chromatography (DHPLC). We identified no obvious pathogenic changes in the coding sequences or splice sites of these genes. Although the study is limited by the small number and heterogeneity of screened patients, and the reduced sensitivity of DHPLC to detect homozygous changes or hemizygous deletions leading to a dosage effect, the findings suggests that neither genes are a common genetic contributor to motor neuron disorders in the human population.

**8. The congenital myasthenic syndromes**

D Beeson
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The congenital myasthenic syndromes (CMS) are rare inherited disorders of neuromuscular transmission, characterised by fatigable muscle weakness. The underlying genetic defects are diverse, as are the pathogenic consequences of dysfunction. CMS account for only around 4% of all myasthenias, but this number is increasing with improved diagnosis. Typically patients present soon after birth with feeding difficulties, a weak cry, and more seriously, difficulties in breathing requiring resuscitation, although some syndromes may not manifest until childhood or even adulthood. Mutations in 11 different genes have now been demonstrated to cause CMS, the latest of which is Dok-7.
Dok-7 is essential for neuromuscular synaptogensis and its interaction with MuSK appears to be crucial for this process. Patients with DOK7 CMS have small, simplified NMJs but the function of the AChR itself appears largely unaffected. Although the interaction of Dok-7 with MuSK is essential for the formation and maintenance of the neuromuscular junction, key components of the downstream pathway that results in AChR clustering have yet to be defined. We have identified a series of differing DOK7 mutations underlying CMS. The study of these mutations is helping elucidate the underlying pathogenic mechanism of DOK7 mutations and the role of Dok-7 in the AChR clustering pathway. Thus, the diversity of affected genes in CMS and the different pathogenic mechanisms of mutations within these genes is proving highly informative for understanding synaptic dysfunction. This is true not only for the AChR itself but also for molecules involved in forming and maintaining the neuromuscular synapse.

**9. A survey of human alpha-dystrobrevin isoforms reveals substantial diversity not present in mouse**

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Alpha-dystrobrevin is one of two paralogous proteins found to heterodimerise with dystrophins in vertebrate animals. As a distant relative of dystrophin itself, it serves to recruit members of the syntrophin family of adaptor protein to the dystrophin complex. The vertebrate alpha-dystrobrevin gene generates a complex array of protein isoforms by a mixture of alternative promoter use, alternative splicing and alternative last exon use. The resulting proteins vary in their abilities to bind members of the dystrophin and syntrophin families, and it is thought that this serves as a means to generate dystrophin/dystrobrevin complexes of specialised stoichiometry and function. Although alpha-dystrobrevin has mostly been studied in mouse, we show here that mice and rats have a specifically reduced repertoire of isoforms when compared to most other mammals, including humans. We present here a qualitative and quantitative description of alpha-dystrobrevin isoforms (including several novel isoforms) in a range of human tissues and discuss the functional implications of novel isoforms, tissue-specific differences and the limitations of the mouse as a model for studying alpha-dystrobrevin function.

**10. Molecular analysis of a family co-segregating myotonic dystrophy type 1 and Charcot-Marie-Tooth disease**

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Myotonic dystrophy type 1 (DM1) is a mutisystemic disorder associated with an expansion of a (CTG)n repeat within the 3'-UTR of the DMPK gene (19q13.3). Charcot-Marie-Tooth disease (CMT) is a genetically heterogeneous hereditary motor and sensory neuropathy of the peripheral nervous system. We are currently investigating the molecular lesion in a very unusual three-generation family in which all the patients co-segregate both DM1 and CMT (LOD score = 7.03). Southern blot analysis of restriction digested genomic DNA has revealed a fragment equivalent to a small CTG expansion (~200-400) at the DM1 locus in all patients. However, an expanded allele could not be amplified by PCR. Similarly, repeat primed-PCR revealed an expanded CTG repeat at the 5'-end of the array, but was negative at the 3' end. Our working hypothesis is that a CTG expansion has been accompanied by an additional lesion such as a deletion, insertion and/or rearrangement. Such a novel mutation might modify the expression of DMPK and/or nearby genes and explain the clinical presentation observed in this DM1/CMT family. Genotyping of flanking SNPs have failed to detect loss of heterozygosity in the immediate vicinity, but have revealed that the mutation is found on the classic DM1 haplotype.

**11. LARGE induced hyperglycosylation of alpha-dystroglycan as a therapeutic strategy in secondary dystroglycanopathies**

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Alpha-Dystroglycan (ADG) is a crucial component of the dystrophin-glycoprotein complex that forms a link between the extracellular matrix and the actin associated cytoskeleton. Glycosylation of ADG is essential for its interaction with a number of extracellular matrix proteins. Recently, defects in 6 known or putative glycosyltransferases have been identified as a major cause of muscular dystrophy (secondary dystroglycanopathies). All forms share a common pathological feature of *hypo*glycosylated ADG.

One gene underlying these disorders is LARGE. The overexpression of this enzyme uniquely induces the *hyper*glycosylation of ADG, increasing its affinity for extracellular matrix proteins. Moreover it also restores ADG glycosylation in cell lines from dystroglycanopathy patients, irrespective of their primary genetic defect. Thus, the upregulation of LARGE expression appears to be a plausible therapeutic approach in these disorders. Key to the development of any such strategies will be knowledge of the degree of upregulation required to induce ADG hyperglycosylation.

We have cloned human LARGE into the pEGSH vector that directs the expression of a FLAG tagged fusion protein under the control of a synthetic ecdysone-inducible promoter and establish stable ER-CHO cell lines harbouring this construct. Transcription of LARGE is induced by the addition of ponasterone A (PonA). Treatment with varying concentrations of PonA demonstrates that this system requires levels of 0.1μM and above to induce robust ADG hyperglycosylation, as demonstrated by both western blotting with antibody IIH6 and laminin overlay assays. Quantitative real-time PCR indicates that 0.1μM PonA increases LARGE expression by a factor of 8. We are currently reproducing these experiments in more relevant systems and correlating the real time data with levels of protein expression. If confirmed, our results indicate that only a relatively modest upregulation of LARGE is sufficient to induce ADG hyperglcyosylation and pharmacological approaches could be developed to achieve this.

**12. Beware all clinical researchers: the FDA's tough new requirements for rating scale performance are coming to a study near you soon**

SJ Cano, JP Zajicek, AJ Thompson and JC Hobart
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The US Food and Drug Administration (FDA) are about to publish strict recommendations concerning rating scales used in clinical trials. The European Medicines Evaluation Agency (EMEA) are following suit. Here, we alert clinical researchers in neuromuscular disease to the FDA's core requirements concerning scale performance, and illustrate why they are essential. FDA requires that any rating scale used in clinical trials satisfies minimum statistical requirements for data quality, scaling assumptions, targeting, reliability, validity, and responsiveness. FDA also requires that clinically meaningful content is established through qualitative evaluation. Our research findings support the need for tough quantitative and qualitative requirements. A literature search shows that state of the art clinical trials continue to use scales proven scientifically poor. Our psychometric evaluations of widely used scales shows basic assumptions are often not met. Our evaluations have also identified scales that satisfied statistical tests of adequate performance despite overwhelming qualitative evidence of invalidity. The FDA guidelines have substantial implications for ALL clinical trials of neuromuscular disease using rating scales. There is strong emphasis on both qualitative and quantitative scale evaluations. Researchers in neuromuscular disease must increasingly become familiar with, and incorporate, these methods in their studies.

**13. Differentiation ability in BL10-SJL skeletal myoblasts**

Y-H Chiu, SH Laval, H Lochmüller, V Straub, K Bushby and R Barresi
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Mutations in the dysferlin gene cause limb-girdle muscular dystrophy type 2B, Miyoshi myopathy and distal anterior compartment myopathy, collectively known as dysferlinopathy. The dysferlin protein containing six C2 domains and one transmembrane domain has been implicated in the membrane resealing step of sarcolemma repair in a previous study. However, the mechanism of the disease and the function of dysferlin in this process are still largely unknown. To understand the function of dysferlin in myoblast differentiation, primary cultures and muscle tissues from wild type C57BL10, dysferlin-deficient BL10-SJL and dystrophin-deficient mdx mice were analysed for growth and fusion as well as dysferlin expression. The primary myoblasts generated from BL10-SJL have a similar division rate and fusion index compared to C57BL10 and mdx mice. In C57BL10 and mdx, dysferlin is expressed at the sites of membrane fusion and cell membrane after differentiation. Surprisingly, although dysferlin was hardly detected in BL10-SJL adult muscles, 20-30% of wild type levels of the dysferlin protein was observed in BL10-SJL neonatal muscle tissue and primary myoblasts generated from neonatal mice by western blot. Further study is necessary to analyse whether dysferlin expressed in BL10-SJL is fully functional. Although there are no differentiation defects in BL10-SJL primary myoblasts generated from neonatal mice, the specific dysferlin expression pattern in C57BL10 and mdx myoculture indicates dysferlin might be involved in myoblast differentiation.

**14. Conservation of a coding function for D4Z4, the repeat sequence associated with facioscapulohumeral muscular dystrophy**

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Facioscapulohumeral muscular dystrophy (FSHD) is caused by a unique genetic rearrangement at HSA 4q35. Unaffected individuals have >12 copies of a tandemly repeated DNA sequence (D4Z4) at this locus, whereas FSHD patients have <10 copies. Each D4Z4 unit is 3.3kb in size and contains an ORF potentially encoding a double homeodomain protein (DUX4). Whilst the mechanism causing FSHD remains elusive, there are two main hypotheses for the effects of D4Z4 deletions. Firstly, that the deletions cause a position effect on nearby genes leading to altered expression and secondly that the mutation perturbs expression of the DUX4 ORF. Recently, we identified D4Z4 homologous sequences in a variety of mammals, including rodents and Afrotheria, demonstrating the conservation of the DUX4 ORF for over 100 million years. We have demonstrated that the mouse *Dux* locus (which is also arranged as a tandem array) is transcribed. Using RT-PCR, we demonstrated that the human D4Z4 locus is also transcribed. We are currently carrying out functional studies of the mouse Dux protein. If the mouse Dux array has a homologous function to human DUX4, investigation of the mouse gene may give insights into the mechanisms underlying FSHD.

**15. Pax7/Pax3 transcriptional activity is required for muscle differentiation and can regulate cell size and proliferation independently of the myogenic program**

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Pax3 and Pax7 are paired-box transcription factors with common and divergent roles in developmental and adult regenerative myogenesis. In satellite cells, the main source of postnatal muscle regeneration, transcriptional activation of Pax7

and Pax3 is associated with different respective phases of quiescence and proliferation, with both factors being down regulated upon differentiation into muscle fibres. We now show that constitutive expression of Pax7 or Pax3 is compatible with muscle differentiation, but expression of dominant negative constructs inhibits expression of both MyoD and myogenin and prevents differentiation from proceeding. Constitutive expression of Pax7 or Pax3 in satellite cells, or in C2C12 myoblasts lacking either protein, results in increased proliferative rate and decreased cell size. Accordingly, expression of dominant-negative constructs leads to slowing of cell division combined with a dramatic increase in cell size. In 3T3 fibroblasts the effects of Pax7 or Pax3 expression or dominant negative inhibition of their transcriptional targets reproduce the effects on cell size and proliferation seen in myogenic cells. These findings show that Pax7/Pax3 transcriptional activity is required for myogenic differentiation, and provide evidence for a novel role in the coordination of cell growth and division that can operate independently of the myogenic program.

**16. Defining progenitor allele length and somatic mosaicism in myotonic dystrophy type 1**

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Myotonic dystrophy type 1 (DM1) is caused by a repeating tract of DNA (CTG) in the DMPK gene. A Southern blot analysis provides an estimate of the disease allele length, however because the mutation tends to expand in an individual, resulting in somatic mosaicism, this estimate only accounts for 50% of the variation in age of onset, a potential indicator of disease severity. Previous work from our lab showed that an estimate of the progenitor allele length (the length at birth) accounts for much more variation in age of onset. There was also a strong relationship between this progenitor allele estimate, and degree of somatic mosaicism, another potential contributor to severity. A limiting factor of that study was that the progenitor allele length was an estimate. For this study, we are directly measuring the inherited progenitor allele in a cohort of adult onset DM1 patients by analyzing Guthrie card blood spots collected at birth. In each individual, inherited alleles will then be compared to the current levels of somatic mosaicism, enabling us to develop objective methods for determining progenitor allele length in DM1 patients. This will provide improved prognostic information and more reliable risk estimates to future generations.

**17. Pronuclear transfer to prevent the transmission of mitochondrial DNA disease**

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Mitochondrial DNA (mtDNA) diseases are an important cause of muscle disease and are often caused by mutations in mitochondrial genes. MtDNA is strictly maternally inherited, so that a woman with mtDNA disease due to a mitochondrial DNA mutation is at significant risk of passing the defect to her children. Therefore, it is important to consider the development of techniques to prevent transmission of mtDNA disease. Experiments in mice have shown that pronuclear transfer can prevent the development of mtDNA disease in the offspring and, following our successful application to the HFEA for a licence to complete this work, we aimed to determine whether this technique could also be performed in human zygotes. We have optimised the technique of pronuclear transfer so that intact karyoplasts containing a pronucleus can be removed from abnormally fertilised human embryos and transferred to another embyro. These embryos show good survival and onward development in culture. Furthermore, mitochondrial DNA analysis has revealed that the level of 'carry over' mtDNA is minimal (2-5%). These data indicate that pronuclear transfer is a feasible strategy to reduce the risk of transmission of mtDNA disease in humans.

**18. Duchenne muscular dystrophy therapy: In Vivo evaluation of small compounds for upregulation of utrophin**

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DMD is caused by mutations in the cytoskeletal protein dystrophin. It is a severe muscle wasting disease which affects 1 in 3000 boys. Strategies based on pharmacological screening, gene and cell-based therapies are currently being pursued in attempts to alleviate the severe phenotype. Advantages of pharmacological screening include ease of systemic delivery of small compounds and the ability to evade immunological and/or toxicity issues. Our strategy of pharmacologically upregulating the dystrophin-related protein utrophin is designed to target the primary defect by restoring sarcolemmal stability in DMD. We have previously demonstrated that 3-4 fold upregulation is sufficient for complete prevention of pathology in transgenic mdx mice. Adenoviral delivery of utrophin demonstrated prevention of pathology in the GRMD dog model. Compounds (~7000) selected for structural diversity by Summit PLC were screened in a high throughput utrophin transcriptional upregulation assay. Several lead compounds identified from this screen have been assessed for their ability to alleviate muscle symptoms in vivo in mdx mice by in-depth analyses including utrophin localisation, expression levels, and

improvement in muscle physiology. Dosing regime is being optimised and medicinal chemistry is used in parallel to improve potency, solubility and low toxicity.

**19. Non-specific over-expression of utrophin in a variety of neuromuscular disorders including limb girdle muscular dystrophies and congenital myopathies**

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Utrophin is a protein that has considerable sequence homology with dystrophin and is encoded by a gene on chromosome 6. It is present with dystrophin on the sarcolemma of foetal and regenerating muscle fibres. In normal mature muscle it is confined to blood vessels, nerves, and neuromuscular and myotendinous junctions. In Duchenne and Becker muscular dystrophy sarcolemmal utrophin re-appears on mature fibres. It is now apparent that this is not a specific phenomenon and varying degrees of sarcolemmal utrophin can be seen in a variety of neuromuscular disorders, including cases with mutations in the genes encoding sarcoglycans, calpain3, the fukutin-related protein and the ryanodine receptor 1. In addition, it can be seen in some neonates with undefined neuromuscular and metabolic conditions and on muscle fibres adjacent to some tumours. These observations illustrate that although the highest levels are seen in Duchenne muscular dystrophy the over-expression of sarcolemmal utrophin is a non-specific phenomenon, that is not only related to the absence of dystrophin. Understanding the reason for utrophin over-expression could be relevant to the pathogenesis of several neuromuscular disorders.

**20. Chloride channel myotonia: exon 8 hot-spot for dominant-negative interactions**

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Myotonia congenita (MC) is an ion channelopathy caused by mutations in CLCN1, which encodes the skeletal muscle voltage-gated chloride channel. We undertook a clinical, genetic and molecular expression study based upon a large cohort of over 300 UK patients. In an initial cohort of 22 families, we sequenced the DNA of the entire coding region of CLCN1 allowing us to undertake a detailed genotype-phenotype correlation study. Generalized muscle hypertrophy, transien weakness and depressed tendon reflexes occurred more frequently in recessive than dominant MC. Mild cold exacerbation and significant muscle pain were equally common features in dominant and recessive cases. Four newly identified dominant mutations clustered in exon 8, which codes for a highly conserved region of predicted interaction between CLC-1 monomers. Based upon these findings we devised an exon hierarchy analysis strategy and applied this to a second cohort of 303 cases. We achieved a genetic diagnosis in 36%. Interestingly, 40 subjects had dominant exon 8 mutations. In total 48 individuals (from 34 families) in cohort 1 and 2 were found to harbour dominant mutations (30% of mutation positive families). We identified 23 new disease causing mutations, confirming the high degree of genetic heterogeneity associated with MC. We propose that exon 8 of CLCN1 is a hot-spot for dominant mutations. Our molecular expression studies of the new exon 8 mutations indicate that this region of the chloride channel has an important role in dominant negative interactions between the two chloride channel monomers.

**21. MRI in Duchenne muscular dystrophy: quantification of fat infiltration and gadolinium uptake using whole-muscle regions of interest**

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This study aimed to evaluate the use of signal intensity for whole-muscle regions of interest (ROIs) for the quantitative investigation of fat infiltration in Duchenne muscle. Muscle free water, and its change with exercise, were investigated using gadolinium contrast and T2 measurements. 11 ambulant boys with DMD (6.6 - 9.9 years) and 6 healthy adult males (29.6 - 35.3 years) were imaged at 3T. Boys were scanned before and four days after stepping exercise and adults four days after stepping exercise. Axial T1-weighted and pre- and post-gadolinium contrast (Omniscan®) images of the calves, thighs and pelvis were obtained, plus a T2 measurement sequence for the thighs. ROIs were drawn using a desktop programme to define muscles at mid-calf, thigh and pelvis. On T1-w images, median signal intensities of DMD muscle were greater than control values, reaching significance for 6/9 muscles studied. Contrast uptake was significantly greater in DMD boys than controls for all muscles studied, except for the lateral gastrocnemii. T2 values for DMD muscle were significantly higher than control. The DMD tibialis anterior showed significantly increased contrast uptake post-exercise. This analytical method can be used to quantify fat infiltration, contrast uptake and T2 values in normal and dystrophic muscle.
This work is supported by the Muscular Dystrophy Campaign with funding from the Big Lottery Fund.

**22. Dominant and recessive mutations in GDAP1 in a small cohort.**
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Mutations in ganglioside-induced differentiation-associated protein 1 (GDAP1) have been identified as a cause of CMT4A and CMT2K. Mutations in GDAP1 can be associated with both axonal and demyelinating phenotypes. The majority of mutations reported so far have been recessive but a minority have been dominantly inherited.
We have sequenced the coding region of GDAP1 in 43 individuals. The following mutations were detected in 4 affected individuals;
1) c.358C>T;p.Arg120Trp which has previously been reported to be dominant.
2) c.[716T>A]+[716T>A];p.[Leu239His]+ [Leu239His] and
3) c.[840delC]+[840delC];p.[Tyr280fs]+[Tyr280fs] in both of these cases the unaffected parents were heterozygous for the mutations and 4) c.399G>A; p.Met133Ile. This mutation has not been seen in controls nor has it been reported in the literature. Its mode of inheritance and pathogenicity is still to be determined.

**23. Mutation analysis in MFN2 demonstrates high locus mutability and mutation hotspot.**
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We have sequenced the coding region of MFN2 in 217 individuals. 22 mutations were detected, of which 13 had not previously been reported. Other affected relatives were available for analysis in 2/13 families; in both, the mutation segregated with the disease phenotype and is likely to be pathogenic. Parental samples were available for 5 probands with no family history of CMT; 4 mutations were shown to be de novo, namely c.280C>T; p.Arg94Trp (2 cases), c.1090C>T; p.Arg364Trp (1 case) and c.754T>A; p.Ser249Thr (1 case). Interestingly, 3 of the 4 de novo mutations have previously been described in the literature and/or detected in other samples referred to this laboratory. Our data demonstrate that the c.280C>T; p.Arg94Trp mutation is a common cause of CMT2 (4/22 samples) and identifies a mutation hotspot in MFN2.
A further 9 individuals had intronic or silent changes not seen in 206 control chromosomes; the clinical significance of these remains to be established.

**24. A novel mutation in the thymidine phosphorylase gene of a patient with mitochondrial neurogastrointestinal encephalomyopathy**
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We report a 22-year-old woman who presented with a 15-year history of recurrent episodes of diarrhoea, vomiting and abdominal pain. She had bilateral ptosis, partial external ophthalmoplegia and sensorineural hearing impairment. There was generalised muscle wasting and weakness in the limbs. Tendon reflexes were absent. Thymidine and deoxyuridine were detected in her plasma, and thymidine phosphorylase (TP) activity was absent in leucocytes and platelets, confirming the diagnosis of mitochondrial neurogastrointestinal encephalomyopathy (MNGIE). Sequencing of the TP gene of the patient and her parents indicated that the patient was compound heterozygous for a previously reported mutation at the 3′ splice site of intron 2 and a novel mutation at the 5′ splice site of intron 9. The novel mutation causes skipping of exon 9, but leaves the reading frame intact. TP protein was, however, not expressed in the patient's fibroblasts. TP deficiency perturbs mitochondrial nucleotide pools, with resulting secondary mitochondrial DNA (mtDNA) abnormalities. The patient's muscle biopsy showed cytochrome oxidase negative fibres. Multiple mtDNA deletions were found in the muscle sample. The patient's fibroblast culture showed a progressive loss of mitochondrial protein expression. The fibroblasts provide a cellular model, which may be used to test the efficacy of potential treatments.

**25. Investigating mitochondrial dysfunction in the Collagen VI-related disorders**
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**Objective:** Mutations in COL6A1, COL6A2 and COL6A3, the genes which encode the extracellular matrix component collagen VI, lead to Bethlem myopathy (BM) and Ullrich Congenital Muscular Dystrophy (UCMD). Despite the extremely mild phenotype of the Col6A1 null mouse, a mitochondrial defect has been demonstrated which was linked to dysregulation of the mitochondrial permeability transition pore (PTP) opening. The same dysregulation has since been demonstrated in UCMD cells in culture, provoking a move towards clinical trials using cyclosporine A, an inhibitor of PTP opening.
**Methods:** We appraised PTP dysregulation by the TMRM assay. TMRM is a fluorescent probe which, as a lipophilic cation, accumulates in polarized mitochondria in proportion to their transmembrane potential. Aberrant PTP opening is signalled by a reduction in TMRM intensity over time.
**Results:** A study of nine UCMD and BM patients showed that PTP dysregulation is rarely seen in fibroblast cell cultures, however in two patients for whom myoblast cultures were available the PTP dysregulation was seen in the myoblast cultures

but not the corresponding fibroblasts. On the other hand, PTP dysregulation is not unique to UCMD myoblasts, although not shared by myoblasts from patients with overlapping conditions. In addition, this defect can be rescued by extracellular matrix constituents other than collagen VI.
**Conclusions:** Abnormality of PTP opening is a non-specific apoptotic phenotype in cell culture and can be rescued by a number of extracellular matrix constituents. We believe that PTP dysregulation is not the major cause of pathology in collagen VI disease.

**26. Functional characterization of the low affinity neurotrophin p75$^{NTR}$ expressing subpopulation of dorsal root ganglion (DRG) neurons**

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Sensory neurons are highly heterogeneous. They can be differentiated on the basis of their functional properties as well as by the expression of growth factor receptors such as p75NTR. This study aims at functionally characterizing the subset marker suitability of anti-p75NTR. Acutely dissociated neurons from the DRG of adult, male, C56/B6 mice were vitally stained with an TRITC-conjugated antibody directed against the extracellular domain of p75NTR antibody (a kind gift of Gipi Schiavo, CRUK) and characterized using Fura calcium imaging in responses to the TRP channel agonists 1μM capsaicin (TRPV1), 250μM menthol (TRPM8) and 100μM mustard oil (TRPA1). The anti-p75NTR labeled 21% of the total neuronal population. Anti-p75NTR staining appeared in both small and large diameter neurons although the brightest staining intensity was generally found in large diameter sensory neurons. Of all anti-p75NTR positive cells, approximately 30% were capsaicin-sensitive, 10% were sensitive to mustard oil, and 6% to menthol indicating that nociceptors as well as thermoreceptors express p75NTR in adult sensory neurons. In conclusion, this study shows that is feasible to use fluorescently tagged antibodies to identify specific subpopulations of sensory neurons and to characterize the functional properties of neurons expressing p75NTR.

**27. Zebrafish knock-down model for muscleblind-like 2**

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Muscleblind-like (MBNL) is a family of three zinc finger proteins which have been implicated in the pathogenesis of Myotonic Dystrophy (DM). Transcribed repeats are retained within the nuclei of DM cells, appearing as foci associated with MBNL1, 2 and 3. It has been suggested that the sequestration of MBNL proteins by repeat expansion transcripts plays a key role in determining DM pathophysiology, including splicing abnormalities.
We have examined the in vivo role of mbnl2 using a loss-of-function approach. We have showed that introducing mbnl2 translation-blocking antisense molecules (morpholinos) in zebrafish embryos results in a dose-dependent specific phenotype, which consists of eye, brain and somite defects, as well as abnormalities of cardiac structure and function. The movements of mbnl2-morphants are restricted and uncoordinated, with exhaustion of avoidance response. Knockdown of zebrafish mbnl2 produces splicing abnormalities and muscle defects, similar to those observed in DM. We were able to rescue this phenotype by the introduction of either the zebrafish or the human version of MBNL2.
The above results indicate that the sequestration of MBNL2 is important and must be considered when developing therapeutic approaches to DM. Additionally, a broader understanding scope of mbnl2 function and relevance during development was derived from our studies.

**28. Ullrich congenital muscular dystrophy fibroblasts and their adhesion to extracellular matrix proteins**

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Ullrich congenital muscular dystrophy (UCMD) is a neuromuscular disorder due to mutations in the three genes encoding for collagen VI chains. UCMD patients have either a complete absence of collagen VI from the muscle and skin or a partial reduction sometimes limited to the muscle basal lamina. In addition to muscle weakness, UCMD patients also have lax ligaments , follicular hyperkeratosis and impaired scarring. It is not fully known yet how the absence of collagen VI leads to the muscle pathology but one contributing factor may be the disruption of the connection between the muscle basal lamina and the extracellular matrix. Even less is known about the pathomechanism underlying the skin phenotype. Taking into consideration the impaired production and deposition of collagen VI in skin fibroblasts cultures from UCMD patients, we investigated the adhesion of patients' fibroblasts (n = 7) to various ECM substrates using an ELISA system. Our results indicate that UCMD fibroblasts have different adhesion properties compared to control fibroblasts. We found a reduction in the adhesion to vitronectin in a patient with COL6A1 mutations and severely reduced collagen VI. In contrast we found enhanced adhesion to tenascin in two patients with partial collagen VI deficiency and with the same heterozygous mutation.

**29. A comparative study of α-dystroglycan glycosylation in dystroglycanopathies indicates differences based on the primary genetic defect**

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Hypoglycosylation of α-dystroglycan underpins a subgroup of muscular dystrophies. Their severity ranges from congenital onset of weakness, severe brain malformations and death in the perinatal period, to mild weakness in adulthood. Mutations in 6 genes have been identified ; three of these (POMT1, POMT2 and POMGnT1) encode for glycosyltransferases catalysing one of the steps in the mannosylation of α-dystroglycan but the function of the other three (Fukutin; FKRP and LARGE) is unknown. The pathological hallmark in these patients is a reduction of immunolabelling in muscle with antibodies to glycosylated epitopes on α-dystroglycan. If the common pathway of all these conditions is the hypoglycosyation of α-dystroglycan , there should be a correlation between the clinical severity and the extent of hypoglycosylation. While this was the case for patients with mutations in POMT1, POMT2, POMGnT1, FKRP and LARGE genes, we found patients with mild limb girdle muscular dystrophy phenotypes without brain involvement and mutations in fukutin who had profound depletion of glycosylated α-dystroglycan. This observation might indicate that factors other than the hypoglycosylation might determine the clinical severity in patients with fukutin mutations, indirectly suggesting that there are differences between the way α-dystroglycan is glycosylated in these conditions.

**30. Down regulation of ColQ by RNA interferences as an alternative therapy in myasthenia**

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AChE is localised and concentrated at the NMJ by the anchoring protein ColQ. It functions to end signal transmission by cleaving the neurotransmitter ACh; either before it reaches the AChR or as it dissociates. Disorders of the NMJ may be either genetic or autoimmune, and often result in a loss of signal reaching the muscle. AChE is the target for one current therapy in myasthenia. AChE inhibitors aim to increase the amount of time ACh is present at the NMJ, enhancing neuromuscular transmission. The aim is to increase neuromuscular transmission using siRNAs targeting ColQ mRNA. Partial loss of the ColQ associated forms of AChE will enhance neuromuscular transmission.
A reporter construct was produced by fusing ColQ cDNA to EGFP, allowing ColQ expression to be followed by fluorescence. The activity of siRNAs targeting ColQ mRNA was investigated in HEK TSA cells. Cells were transiently transfected with siRNAs and ColQ-EGFP. The activity was followed by fluorescence, western blotting and real time PCR. ColQ (without EGFP) has also been expressed along with AChE in HEK TSA cells allowing the effect on ColQ associated forms of AChE to be followed using sucrose density gradients and Ellman AChE assays.
Three siRNAs were designed against ColQ mRNA, targeting different positions along its length. Down regulation of ColQ mRNA was efficient using two siRNAs. In their presence fluorescence was reduced to 15% of controls. Results from western blotts show an almost complete loss of ColQ expression. Real time PCR reveals a loss of mRNA levels to 30% of controls. siRNAs against ColQ mRNA effectively down regulate expression in HEK TSA cells. Our future work will look at their effects in the AChR deficient mouse model of myasthenia.

**31. Immune deficiency in a mouse model of CMT4D**

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Hereditary Motor and Sensory Neuropathy Lom (HMSNL) also called Charcot-Marie-Tooth Disease 4D (CMT4D) is a severe, disabling peripheral nerve disease in which initial demyelination due to a mutation in n-myc downstream regulating gene 1 (NDRG1) is followed by extensive axonal loss. In this disease, as in other demyelinating diseases, the morbidity is mainly due to the secondary axonal involvement. In our two mouse models, ndrg1 protein is completely absent in one and severely reduced in the other. Initially peripheral nerve myelination progresses normally in both strains, but soon after all fibres are myelinated (14-18 days) occasional demyelinated fibres may be found. At first, only occasional, scattered demyelinated axons are present and the incidence of demyelination remains low until about 4 weeks of age when it increases sharply. Demyelination predominantly affects the larger diameter fibres. Concurrently, the nerve conduction velocities fall steeply to 6 weeks age of age and then stabilize at about 10m/s. After about 30 weeks of age the nerves appear more stable with fewer actively demyelinated fibres or cellular infiltration. Electron microscopy of nerve roots from 2 and 3 week old animals show associated macrophages within the myelin sheath. This suggests that a secondary immune response generates a positive feedback mechanism causing progressive demyelination, and may have therapeutic implications for patients with CMT.

**32. Dysferlin in muscle regeneration**

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Mutations in the dysferlin gene cause LGMD2B, Miyoshi Myopathy and distal anterior compartment myopathy. Dysferlin, a 230 kDa transmembrane protein with six C2 domains, plays a role in the process of membrane maintenance and repair. In mature skeletal muscle dysferlin is mainly located at the sarcolemma with only a minor proportion being associated with the T-tubule system. This pattern of subcellular distribution is altered during the process of regeneration where dysferlin is predominantly located in the cytoplasm. This phenomenon can be observed in regenerating myofibres across different forms of muscular dystrophies and so far is unexplained. We have previously shown that dysferlin fails to localise to the sarcolemma but mainly associates with the T-tubule system of C2C12 myotubes, an in vitro model of myogenesis. To gain further insight into dysferlin's localisation and role in muscle regeneration we injected female Wistar rats with notexin, a snake venom which causes myofibre breakdown leaving satellite cells intact. Muscle was collected and analysed at different time points in the regenerative process. Detection via immunofluorescence demonstrated that 1) there is high abundance of dysferlin at the sites of contact between satellite cells and early myotubes, 2) the localisation is mostly cytoplasmic in longitudinal orientated structures in early regeneration, 3) there is almost complete colocalisation of dysferlin and the developing T-tubule system. These results corroborate previous in vitro findings of dysferlin expression at sites of myoblast/myotube fusion. More importantly, we demonstrate colocalisation of dysferlin and the T-tubule system during the process of regeneration in vivo, which strongly indicates an involvement of dysferlin in T-tubulogenesis. Taken together, our data indicate a specific role of dysferlin in the process of muscle regeneration, most likely in the organisation and fusion of membrane compartments.

**33. *In-vivo* electromyography demonstrates improvement in neuromuscular transmission in mouse models of congenital myasthenic syndromes following conventional treatment**

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Congenital myasthenic syndromes (CMS) are inheritable disorders of neuromuscular transmission. We have developed mouse models of two CMS disorders – acetylcholine receptor (AChR) deficiency and Slow Channel Syndrome (SCS) in order to investigate novel therapies. These preliminary studies aim to develop the techniques of in-vivo electromyography (EMG) and validate the models using existing therapies.

Recordings of the compound muscle action potential (CMAP) with repetitive nerve stimulation (RNS) at 5Hz and stimulated single fibre electromyography (s-SFEMG) at 10Hz were made in the gastrocnemius muscle of mouse models of AChR deficiency and SCS. Both models demonstrated CMAP decrement (5th versus 1st, peak to peak amplitude) (19.4 ± 6.0 %, n = 9 and 29.6 ± 5.0 %, n = 4 respectively) and raised MCD (21.0 ± 5.6 μs, n = 4; 18.2 ± 4.5 μs, n = 3) versus controls (3.1 ± 1.4 % and 12.2 ± 0.7 μs, n = 6).

Mice treated orally for 4 weeks were then compared to untreated controls. In preliminary results, AChR deficiency mic treated with pyridostigmine (5mg/kg/day, n = 3), 3,4 diaminopyridine (1mg/kg/day, n = 4) and the combination of both (n = 3) had reduced fatigable weakness (inverted screen test 195 ± 69 s; 280 ± 24 s and 256 ± 53 s respectively), lesser CMAP decrement (1.2 ± 4.4 %; 4.7 ± 0.4 % and 2.9 ± 0.8 %) and equal MCD (18.5 ± 4.6 μs; 14.9 ± 1.9 ms and 20.8 ± 2.9 μs) versus controls (49 ± 16 s; 8.5 ± 11 % and 21.0 ± 5.6 μs, n = 4). In the first SCS mice treated with fluoxetine (20mg/kg/day, n = 2), treatment resulted in less fatigable weakness (224 s), less decrement (4.3 %) and lower MCD (14.4 μs) than controls (129 ± 19 s; 27.0 ± 8.0 %; 19.7 ± 2.2 μs, n = 3).

These early results indicate that the use of in vivo EMG in our animal models will provide a suitable system for investigating novel therapies.

**34. Charcot Marie Tooth type 4C caused by mutation of KIAA1985 gene: report of 6 families with variable phenotype**

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Charcot Marie Tooth (CMT) disease is a heterogeneous group of inherited motor and sensory neuropathies. CMT4C is an autosomal recessive (AR) demyelinating neuropathy, characterized by early onset, frequent and severe scoliosis and distinct Schwann cell pathology. The locus responsible for CMT4C was assigned to the chromosome 5q23-q33 region by homozygosity mapping and the SH3TC2 (KIAA1985) gene was identified mainly in families around the Mediterranean basin but also in Dutch and European Gypsies. We report five English families with AR demyelinating CMT due to a homozygous mutation in the SH3TC2 gene. All families were homozygous for the Arg954Stop mutation. A sixth family had two compound heterozygous mutations, a Arg954Stop and a Glu657Lys. There was significant phenotypic variability between these families. Some cases presented with a severe early onset neuropathy with respiratory and cranial nerve involvement whereas some other cases presented with mild scoliosis and foot deformity. The phenotype varied also in the same kinship. One patient had a 20 year history of a presumed inflammatory neuropathy that was superimposed onto the hereditary neuropathy.

**35. Randomised double blind placebo controlled trial of long term ascorbic acid treatment in Charcot-Marie-Tooth disease type 1A (CMT-TRAUK: CMT-TRial with Ascorbic acid United Kingdom)**

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Charcot-Marie-Tooth (CMT) type 1A associated with the peripheral myelin protein 22 (PMP22) gene duplication is one of the most common hereditary neuropathies. Recently Ascorbic Acid (AA) was shown to be effective in treating transgenic mice overexpressing PMP22. We are conducting a phase III randomized, double blind, placebo-controlled parallel group trial involving 50 patients with CMT1A. The primary objective of the trial is to assess the efficacy of chronic treatment with AA in patients with CMT1A. Secondary aims are to develop and validate an evaluation protocol that is suitable for future trials in CMT disease. Treatment consists of two-year oral AA (1500 mg/day) or placebo. The primary outcome measure will be an improvement in the Charcot-Marie-Tooth Neuropathy Score (CMTNS). Secondary efficacy endpoints are changes in distal maximum voluntary isometric contraction; 10-meter timed walking; 9-hole-peg test; Overall Neuropathy Limitations Scale; pain and fatigue VAS; health-related quality of life (SF-36); nerve conduction studies. Also small fibre involvement and pain characteristics are studied using thermal thresholds, contact heat evoked potentials and pain questionnaires. The recruitment have started in March 2007. Between March-September 2007, a total of 62 patients have been screened. Recruitment is now finished with 50 patients randomized.

**36. Identification and characterization of a candidate gene for ostes, a novel mouse mutant showing complex neuromuscular phenotypes**

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We have exploited N-ethyl N-nitrosourea (ENU) mutagenesis to identify new mouse models and thus novel genes involved in muscular atrophy. Ostes is a novel chemically-induced genetic mouse mutant which displays muscle atrophy, abnormal muscular innervation and delayed axonal regeneration, tremors and growth retardation.
We genetically mapped the ostes mutation to a 0.8Mb region on distal mouse chromosome 8. Intriguingly, no coding mutation was revealed upon sequence analysis of all 9 genes in the region. Expression analysis of wildtype and ostes/ostes muscle did not show any difference in candidate genes. We identified Pkd1l2 as a candidate gene. Western blot analysis revealed a >200kDa Pkd1l2-specific band that was overexpressed/overaccumulated in skeletal muscle from ostes/ostes mice and also from transgenic mice carrying a BAC that contained the full-length Pkd1l2. Subsequent genetic crosses showed that a) the BAC transgene rescued the ostes/ostes phenotype and b) homozygous BAC transgenics displayed severe muscle wasting, periodic paralysis and juvenile lethality.
Our results indicate that mutation or misregulation of the Pkd1l2 protein is associated with neuromuscular defects in two mouse mutants. We suggest that Pkd1l2 is a crucial regulator of complex neuromuscular functions and a candidate gene for uncharacterised forms of neuromuscular disease.

**37. Nondystrophic myotonias: genotype-phenotype correlation and longitudinal study**

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The non-dystrophic myotonias are rare episodic neuromuscular disorders. No large scale genotype-phenotype correlation or natural history studies have been performed. As members of the CINCH group (Consortium for Clinical Investigation of Neurologic Channelopathies) we are involved at the MRC Centre for Translational Research of Neuromuscular Disorders, UCL Institute of Neurology, in an international collaboration of centres carrying out a natural history trial of the non-dystrophic myotonias.
Participants are enrolled for three years into the study and information is collected on symptom frequency and severity, presence and degree of myotonia, muscle power, electrophysiological data and quality of life. Within the time-frame of the trial genetic sequencing of all known causative genes is undertaken. This should allow for accurate phenotype-genotype correlation and insight into the natural progression of these rare diseases. It is expected this trial will also identify suitable end-points for future treatment trials.
At our centre we have been able to illustrate already that EMG is accurate in predicting causative gene. To date, 78% of our participants have been genotyped. Physiotherapy has confirmed proximal myopathy as a common finding in NDM occurring in 14/18 participants although 8/18 also had distal weakness, a feature not well recognised. Symptom review is also providing new insights, 16/18 participants reported myalgia which has not previously been a commonly considered symptom in NDM.

**38. Sodium channel [SCN4A] periodic paralysis with severe myopathy-possible role of a pore leak current**

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Mild myopathy is common in paients with periodic paralysis although its precise molecular pathophysiology remains undetermined. We have identified a family with unusually severe myopathy and periodic paralysis. Symptoms start in the first years of life with periods of paralysis often lasting hours to days. This is followed by lesser but still significant weakness for weeks or even months. Following such prolonged episodes as this muscle power does not return to full strength and a permanent myopathy has developed. The men in the family are particularly severely affected, both requiring wheelchairs from the mid thirties and help with ADLs. Recently we identified a mutation in the SCN4A gene which encodes the skeletal muscle sodium channel in affected individuals. It predicts the substitution of an arginine for a glycine in the S4 segment of domain II of the channel. Substitutions of adjacent arginine residues in this segment, which acts as the "voltage sensor" for the channel, lead not only to a prolonged flow of sodium through the main channel but also a "pore leak" when the main channel is closed. It is possible that pore-leak mutations may associate with more severe myodegeneration than non-pore leak mutations. We describe the genetic and molecular expression findings in this family and discuss the role of pore leak currents and muscle degeneration.

**39. Morphological, stem cell and myosin abnormalities in the cav-3-/- and mdx dystrophic embryo reveal an embryonic basis for muscular dystrophy**

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Examination of embryonic myogenesis of two distinct but functionally related, skeletal muscle dystrophy mutants (mdx and cav-3-/-) establishes for the first time that key elements of the pathology of DMD and LGMD type 1C originate in disruption of the embryonic cardiac and skeletal muscle patterning processes. Novel elements of the phenotype suggest compromised stem cell function has an important role in the aetiology and severity of both diseases. Hyperproliferation and apoptosis in myf-5+ embryonic myoblasts and attrition of the pax-7+ stem cell population in situ occurs earlier in mdx (E11.5) than cav-3-/- (E15.5) but results in depletion of total pax-7 protein respectively in mdx and cav-3-/- embryos by 15% and 60%. mdx embryos have several fold elevation of caveolin-3 which may contribute to the eitiology of mdx/DMD pathologies with reciprocity with cav-3-/-. In mdx, myotube numbers are reduced and myotubes misaligned, hypotrophic and branching. The more restricted phenotype of cav-3-/- comprises excess, malformed hypertrophic myotubes and 2-fold increase in myonuclei. Myosin heavy chain (MyHC) content is disrupted in both mutants as is timing of secondary myogenesis. We conclude that caveolin-3 and dystrophin have important embryonic roles and act together to ensure the normal progression of embryonic muscle formation. MD pathology originates in embryonic myogenesis through early failure of the DGC and disruption of the emerging skeletal muscle stem cell population.

**40. Imaging mRNAs involved in myotonic dystrophy type 1 using atomic force microscopy**

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Atomic force microscopy (AFM) is a recent form of microscopy that has now become an established imaging tool within the nanotechnology world. Unlike conventional optical microscopy, AFM uses a micrometre-sized tip probe that is allowed to scan over a flat surface where the sample of interest is dispersed. As the probe scans the surface, the change of height caused by the encounter of the sample is detected through a laser detection system and the data collected are further processed to re-create an image of the sample. In the past decade, AFM has been successfully used for imaging biological samples such as nucleic acid, proteins and their subsequent complexes.
In this study, AFM has been used to image mutant DM mRNAs molecules that contain an over-expanded number of CUG repeats (140 repeats) and also their interaction with MBNL1 proteins. Results show that DM mRNAs containing 140 CUG repeats form a very stable double-stranded RNA hairpin that can be easily imaged in air using AFM. We have also imaged the interaction of these mRNA with MBNL1 proteins and a distinctive motif of the binding to the CUG hairpin is recognisable. These results are the very first images showing MBNL1 proteins binding to mutant DM mRNAs. These encouraging results show that AFM could later be used as system to understand better the molecular mechanisms underlying Myotonic Dystrophy as well as understanding the mechanism of action of potential therapies.

**41. Characterisation of a satellite cell derived cell-line from the H-2Kb-tsA58 immortomouse.**

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In order to optimise a new non-viral gene therapy system (Synthegene) in a muscle stem cell model, a new immortalised satellite cell derived cell-line was developed from the H-2Kb-tsA58 immortomouse.
The satellite-derived cell line was characterised firstly, to ensure it was a suitable representation of a muscle stem cell and secondly, to define the characteristics of the cells prior to genetic modification. Satellite cell derived clones were prepared from single myofibres from the H-2Kb-tsA58 immortomouse (Jat et al, 1991). The H2K 2B4 clone was chosen for further characterisation as it exhibited low doubling time and the ability to differentiate in vitro. A four-day in vitro differentiation assay showed the expected expression pattern of the myogenic markers: Desmin, Fast/Slow Myosin, Pax 7, MyoD and Myogenin. When transplanted into irradiated mdx nu /nu mouse muscle, the cells were able generate muscle which remained for at least six months, as well as contribute to the satellite cell population.
The clone can be transfected by Nucleofection or transduced by retrovirus with both methods achieving approximately 50% of the cells expressing the GFP transgene, whilst retaining the ability to differentiate into myotubes in vitro.
Overall the H2K 2B4 cell line has shown to be a reliable cell line which would ideal to be used as a muscle stem cell model for the optimisation of the Synthegene system.

**42. DOK7 mutations in congenital myasthenic syndromes: clinical features and implications for therapy**

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Recently, a novel neuromuscular junction protein termed Dok-7 has been described. Subsequently, we and others identified mutations in the DOK7 gene as a cause of congenital myasthenic syndromes (CMS). DOK7 mutations have emerged as one of the major genetic defects in CMS: In our cohort 20 CMS patients from 17 independent kinships harboured DOK7 mutations, accounting for about 10% of genetically diagnosed CMS cases.
The clinical picture of CMS with DOK7 mutations is highly variable and differs significantly from CMS caused by mutations in other genes. The age of onset may vary between birth and the third decade. However, most of the patients display a characteristic 'limb-girdle' pattern of weakness with a waddling gait and ptosis, but without ophthalmoparesis. Progressive deterioration of respiratory function is frequent. Patients with DOK7 mutations do not benefit from long-term therapy with esterase inhibitors; some of the patients even worsened. A pilot therapy with ephedrine was started in three of our DOK7 patients and showed an improvement of clinical symptoms with only few side effects. These results could be observed even when therapy started several years after manifestation of the disease, emphasizing the possibility of ephedrine therapy for CMS patients with DOK7 mutations.

**43. Exercise training in patients with mitochondrial myopathy**

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Mitochondrial myopathies are a group of progressive muscle disorders caused by mutations in the mitochondrial genome (mtDNA). Patients can present with a variety of symptoms, however muscle symptoms often predominate leading to severe disability or death. Despite progress in the diagnosis and conservative management of patients with mitochondrial myopathies, there remains a need for effective treatments. The work presented here involves investigating the use of exercise training – both endurance and resistance – as a potential treatment for a genetically-homogeneous group of patients with single, large-scale mtDNA deletions.
We found that 14 weeks of endurance exercise training improved submaximal exercise tolerance and peak capacity for work, oxygen utilisation and skeletal muscle oxygen extraction with no change in the level of deleted mtDNA. Continued training for an additional 14 weeks maintained these beneficial adaptations, whereas the cessation of training resulted in loss of physiological adaptation with no overall change in mutation load.
Similarly, following 14 weeks of resistance exercise training, patients showed improved muscle strength, with a concomitant increase in markers of muscle regeneration. The percentage of cytochrome c oxidase (COX) deficient muscle fibres decreased, with an increased proportion of intermediate staining fibres, suggesting partial restoration of the biochemical defect.

**44. Muscle satellite cell fate choice is influenced by b-catenin**

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Satellite cells are the resident stem cells of adult skeletal muscle. Their population is maintained by self-renewal, and how the choice to differentiate or self-renew is controlled, is central to understanding their function. Here we have explored the role played by β-catenin in satellite cell fate choice. Satellite cells
express β-catenin, which is maintained as they activate and undergo proliferation. Constitutive expression of wild type or stabilised β-catenin resulted in more satellite cells expressing Pax7 without MyoD and so adopting the self-renewal pathway, with fewer satellite cell progeny undergoing myogenic differentiation. Indeed constitutive β-catenin is able to induce Pax7 expression in myogenic cells. Similarly, preventing degradation of endogenous β-catenin by inhibiting GSK3β activity also resulted in more satellite cell progeny expressing Pax7 and not differentiating. Consistent with these observations, it was found that using siRNA to down-regulate β-catenin or a dominant negative version to repress β-catenin transcriptional targets in satellite cells, both augmented differentiation. Together these observations show that β-catenin is able to direct proliferating satellite cells to the self-renewal pathway and away from immediate myogenic differentiation.

**45. Increased sensitivity to oxidative stress in myoblast and fibroblast cultures from amyotrophic lateral sclerosis patients**

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We compared oxidative stress, mitochondrial respiratory chain function and mitochondrial DNA (mtDNA) levels in peripheral tissues, including muscle, from 12 patients with motor neuron disease or amyotrophic lateral sclerosis (ALS) with control samples. Aconitase activity measurements did not indicate increased oxidative damage in muscle, or in cultured myoblasts or fibroblasts from ALS patients. We did, however, find an increased sensitivity to oxidative stress in cell cultures from ALS patients exposed to paraquat compared to control samples. Respiratory chain enzyme activities were normal in muscle and cultures from ALS patients, as were levels of mtDNA in muscle. Rearranged muscle mtDNA species were not detected by Southern blot hybridization in any of the samples and no difference was found between the patient and control group in the number of deleted mtDNA species detected by PCR. Platelet-derived cybrid studies confirmed the absence of a systemic mtDNA abnormality. The increased sensitivity to oxidative stress may become detrimental in the more metabolically active motor neurons of ALS patients. Our study suggests that this altered sensitivity is due to a nuclear rather than a mtDNA abnormality. These findings of systemic biochemical abnormalities may allow the development of a more accessible biomarker of disease activity and response to medication, than the inaccessible motor neurons.

**46. Hemi-atrophy- An unusual phenotype in a manifesting carrier of Duchenne muscular dystrophy**

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We present clinical, muscle biopsy and genetic findings in a female DMD manifesting carrier presenting with marked right hemi-atrophy. We review the literature on the presentations of manifesting DMD carriers

**Case Report**

We studied a 35 year old woman with a 17 year history of progressive wasting and weakness in her right arm and right leg. The musculature in the left arm and leg was normal in appearance and power. Her facial muscles were unaffected. There was no family history.

Investigations: Raised creatine kinase level [900] and myopathic changes on EMG studies. Cardiac evaluation will be presented. MRI of her Brain and Spine were normal. Genetic testing for common mitochondrial mutations [muscle mtDNA –deletions, 3243, 8344, 8993] were negative. The FSHD Ch4 truncation was not detected. Her muscle biopsy demonstrated a significant reduction in dystrophin immuno-staining, suggesting that she was a manifesting carrier of DMD. A screen for deletions, duplications and point mutations in the dystrophin gene revealed a duplication of exons 10 and 11. DNA sequencing did not reveal any point mutations in the dystrophin gene.

Such a marked asymmetrical presentation of a manifesting DMD carrier is unusual. We present the results of her clinical and genetic investigations and explore the possible mechanisms of the asymmetric muscle involvement.

**47. Characterisation of the dysferlin related proteins, myoferlin and FER1L5, potential compensatory proteins of dysferlin**

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The gene encoding dysferlin is mutated in autosomal recessive Miyoshi myopathy, Limb Girdle Muscular Dystrophy type 2B (LGMD2B) and distal anterior compartment myopathy, causing dysferlin deficiency in patient muscle. Dysferlin is a sarcolemmal protein sharing homology with the sperm vesicle fusion protein FER-1, which mediates fusion of intracellular vesicles with the spermatid plasma membrane. Dysferlin has been shown to be involved in sarcolemmal repair through a proposed mechanism that involves fusion of dysferlin containing vesicles to form a "membrane patch" which is added to the membrane disruption site for resealing. We now know that dysferlin is a member of the ferlin protein family structurally similar proteins sharing homology with C. elegans FER-1 and also predicted to be involved in membrane fusion. We have sought the identification of those sharing similar properties and distribution with dysferlin in muscle to assess their validity as potential compensatory molecules of dysferlin.
To date three ferlin proteins, dysferlin, myoferlin and otoferlin have been reported. Through bioinformatic analysis we have identified three novel ferlins, FER1L4, FER1L5 and FER1L6. Homology modeling of all of the ferlin C2 domains and sequence analysis has identified myoferlin and FER1L5 as being the most similar to dysferlin. We have performed the molecular characterisation of the novel FER1L5. We have also examined the relative distribution of myoferlin, FER1L5 and dysferlin in mouse muscle and C2C12 cells by sucrose gradient fractionation. We demonstrate that dysferlin, myoferlin and FER1L5 are present in membrane vesicles which are predominantly low density vesicles similar to other organelles. The sedimentation profiles of myoferlin vesicles appear most similar to dysferlin. To examine membrane repair we have adopted siRN transfection studies. We have successfully generated and characterized myoferlin deficient C2C12 cells. For FER1L5 siRNA knockdown of FER1L5 protein has not been possible but we have performed antibody loading to disrupt protein function. The cellular phenotype of these cells will be discussed.

**48. Treatment with mechanogrowth factor, an IGF-1 splice variant, rescues motoneurons and delays disease progression in the SOD-1G93A mouse model of motor neuron disease**

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Mechanogrowth factor (MGF) is a splice site variant of IGF-1, which has been shown to have myotrophic and neurotrophic effects. There is evidence that MGF may have greater potency than IGF-1. We have investigated the neurotrophic and myotrophic effects of MGF. Here, we examined the potential therapeutic benefits of MGF in the SOD1G93A mouse model of motor neuron disease (MND). Hindlimb muscles of SOD1G93A mice were injected with a mammalian expression plasmid containing the MGF cDNA sequence at 70 days, an early symptomatic stage of the disease. At 120 days, hindlimb muscle force and motor unit survival was assessed by in-vivo physiological recordings. The spinal cord and muscles were subsequently removed and motoneuron survival and muscle histochemistry examined.
By 120 days, the hindlimb muscles of untreated SOD1G93A mice were significantly weaker than normal and, for example, Tibialis Anterior (TA) muscles produced only 10% of the force seen in wild-type littermates. In contrast, in MGF treated SOD1G93A mice, the TA muscles were significantly stronger ($p<0.021$). Moreover, more functional motor units survived in MGF treated SOD1 G93A mice than in their untreated SOD1 G93A littermates. This improvement in motor unit survival was reflected in an increase in motoneuron survival in the spinal cord of MGF treated SOD1 G93A mice compared to untreated SOD1 G93A mice ($p=0.029$). MGF gene transfer to SOD1 G93A mouse skeletal muscle rescues motoneurons and improves muscle function. MGF may have therapeutic potential in ALS, and other neuromuscular disorders.
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**49. Delivery of morpholino antisense oligonucleotide improves muscle force and ameliorates contraction-induced injury in a mouse model of muscular dystrophy**

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Duchenne muscular dystrophy (DMD) is a progressive muscle degenerative disorder resulting from mutations in the dystrophin gene that prevents the synthesis of functional protein. The most commonly used mouse model of DMD is the mdx mouse, a naturally occurring dystrophin mutant with a nonsense mutation in exon 23. A potential therapy consists of using antisense oligonucleotides (AOs) to redirect gene transcript processing in order to bypass the mutations in the dystrophin gene and thereby allowing the production of a truncated but functional dystrophin protein. Recent studies have shown that morpholino based chemistry is particularly promising, since these AOs are non-toxic and highly resistant to nuclease activity thereby improving their potential for remaining active in cells for a prolonged period. In this study, we examined the efficacy of morpholino AOs to produce dystrophin over time in tibialis anterior muscles (TA) of mdx mice and whether this was sufficient to improve physiological function. We found that 4 weeks after a single intramuscular injection of morpholino AO, dystrophin expression was detected in a large number of fibres of the mdx TA muscle, which most

importantly resulted in an improvement in normalised muscle force and also ameliorated the decline in force that occurs in dystrophic muscle following injury-inducing contractions. However, by 8 weeks after administration the number of dystrophin positive fibres had declined, which correlated with the loss of functional benefits. These pre-clinical studies demonstrate the effectiveness of the morpholino based AO in improving the dystrophic phenotype and this approach is now being taken forward to a UK clinical trial in DMD by the MDEX Consortium.

**50. Comparative human mitochondrial genome analysis using the Affymetrix MitoChip and conventional cycle sequencing**

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**Introduction:** Mitochondrial DNA (mtDNA) is a circular molecule of 16,569 kb. The pathology of mitochondrial disease can be due to mutations in either nuclear DNA or mtDNA.
Previously, we have sequenced the entire mitochondrial genome using 45 overlapping PCR fragments. As each fragment should ideally be sequenced in both the forward and reverse direction, this is both a lengthy and costly procedure.
Recently, Affymetrix have launched an updated version of their commercially available resequencing chip for the mitochondrial genome. The MitoChip is an array-based sequencing platform for rapid and high throughput analysis of mtDNA. We aim to evaluate the efficiency of the MitoChip in sequencing mtDNA in its entirety.
**Methods:** A cohort of 24 patients, with mitochondrial disease but without a genetic diagnosis, was identified through referral to the Neurogenetics laboratory at the National Hospital for Neurology, Queen Square, London. Sequence analysis of mtDNA using both conventional sequencing and the MitoChip, and subsequent comparison of both data sets will evaluate the efficiency of the resequencing chip in identifying pathogenic mutations. In addition we will determine the frequency of false positives and regions consistently mis-called by the Affymetrix resequencing software.
**Results and Discussion:** To date, 9 patients have been analysed using the MitoChip. Four mutations have been identified, and subsequently confirmed by conventional sequencing.
Preliminary data shows the MitoChip to be a potentially useful tool in the identification of mtDNA mutations. Completion of the analysis of the cohort by both methods will allow us to fully evaluate the v2 MitoChip, and highlight any potential pitfalls of the method.

**51 How does a dynein mutation slow motor neuron disease?**

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Despite extensive research, the molecular basis of Motor Neuron Disease (MND) remains poorly understood. Genetic forms of MND offer potential insight into this basis. Mutations in superoxide dismutase 1 (SOD1G93A) are responsible for 5-20% of cases of familial MND. This mutation has been modelled in the SOD1G93A transgenic mouse (SOD1G93A mouse) which exhibits motor neuron loss, muscle weakness and premature death. The Loa mouse carries a mutation the gene encoding a subunit of cytoplasmic dynein. This mouse also shows motor neuron dysfunction but heterozygotes have normal lifespan. Crossing Loa and SOD1G93A mice produce offspring of four genotypes – wild-type (+/+), Loa/+, SOD1G93A /+ and Loa/SOD1G93A. Surprisingly, Loa/SOD1G93A mice survive significantly longer than their SOD1G93A /+ littermates, thus the dynein mutation appears to reduce the toxic effects of SOD1 G93A. To compare the dynamics of SOD1G93A between motor neurons from these mice, we transduced neurons with GFP-SOD1G93A using a lentiviral vector. Neurons of these four genotypes show no variation in the distribution of SOD1G93A or its tendency to aggregate. Live cell imaging to asses transport of wild-type and mutant SOD1 within motor neurons is underway. By comparing the behaviour of SOD1 between neurons with different survival characteristics, we aim to gain further insight into its toxic mechanism.

**52. New mutations of the SCN4A gene in patients with paramyotonia**

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The SCN4A gene encodes the voltage gated sodium channel alpha subunit Nav1.4. Dominantly inherited skeletal muscle sodium channelopathies associated with mutations in SCN4A include paramyotonia congenita (PMC), the potassium aggravated myotonias (PAM), hyperkalaemic periodic paralysis, and rarely hypokalaemic periodic paralysis. Typical clinical features of PMC are myotonia exacerbated by exercise and cold exposure which is often followed by weakness. Muscles in the face and upper limbs are primarily affected and onset is usually in the first decade. In contrast myotonic symptoms in PAM are not associated with weakness or cold exposure but with potassium ingestion. We present results of the analysis of 145 individuals referred to our laboratory for genetic testing of PMC and PAM. We screened exons 22 and 24 of the SCN4A gene which contain the mutations that frequently cause PMC. Twelve different SCN4A mutations were identified. Those most commonly observed were Thr1313Met and substitutions for an arginine at position 1448 including a newly identifie

substitution. A second new mutation was detected at Leu1436. Patients with the new mutations displayed a typical PMC phenotype. Gly1306Glu and Gly1306Ala mutations that have been previously reported to cause PAM were unusually associated with cold exacerbation in 2 families.

**53. Paradoxical effects of a CACNA1A mutation on neurotransmitter exocytosis in hippocampal neurons**

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Presynaptic P/Q type calcium channels mediate neurotransmitter release at synapses of the central and peripheral nervous system. Mutations in the underlying CACNA1A gene are associated with paroxysmal neurological disorders such as familial hemiplegic migraine type 1 (FHM). The S218L mutation results in severe FHM together with susceptibility to brain oedema. At neuromuscular junctions this mutation is associated with an increased neurotransmitter release probability in S218L knock-in (KI) mice, when compared to wild-type mice [Kaja et al., submitted]. We have investigated the effects of S218L mutations on the rates of synaptic vesicle exocytosis at individual synapses in cultured hippocampal neurons of the S218L KI strain using the FM dye technique. In contrast to neuromuscular junctions, the effect of S218L mutation on central synapses was complex. Overall we observed an inhibition of evoked synaptic exocytosis in both heterozygous and homozygous S218L mice as compared to wild type mice, mainly due to a 1.7-2.5 fold increase in occurrence of "silent" synapses. However at the same time the number of synapses with high release probability was increased in both heterozygous (1.5 fold) and homozygous (5 fold) mice. Thus a single mutation can lead to opposite effects on neurotransmission in different subsets of central synapses.
Supported by the DFG, MRC and European Commission (Eurohead)

**54. Cell culture models of cytochrome-c oxidase assembly defects**

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We have systematically silenced the expression of the ten nuclear-encoded cytochrome-c oxidase subunits in HeLa cell cultures by RNA interference to clarify the contribution of each subunit to assembly. Silencing resulted in >90% knock-down of the targeted subunits in all cells. Residual cytochrome-c oxidase activity in the silenced cultures mirrored the residual level of the targeted subunit, except for subunit COX6A1-silenced cultures, which showed relatively normal activity. Thus, apart from subunit COX6A1, all nuclear-encoded subunits are required for assembly of a catalytically active enzyme. Immunoblots of blue native gels revealed subassemblies involving subunits MTCO1, COX4I1 and COX5A in control cultures and in subunit COX6A1, COX6B1, COX6C and COX7A2 knock-down cultures, but not in subunit COX4I1, COX5A, COX5B, COX7B, COX7C and COX8A knock-down cultures. Subassemblies involving subunits MTCO2, MTCO3 or COX6C were not detected in any of the cultures. These results suggest that core subunit MTCO1 may form relatively stable subassemblies with subunits COX4I1, COX5A, COX5B, COX7B, COX7C and COX8A in the absence of subunits MTCO2, MTCO3, COX6A1, COX6B1, COX6C and COX7A2. Comparisons of subassembly pattern "fingerprints" present in our cell culture models with those present in patient cells will indicate which step of the assembly pathway is disrupted in patients.

**55. Analysis of mutant DNA polymerase γ in fibroblast cultures from patients with mitochondrial DNA depletion**

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We studied six unrelated children with mitochondrial DNA (mtDNA) depletion. They presented with Leigh syndrome, infantile hepatocerebral mtDNA depletion syndrome or Alpers-Huttenlocher syndrome. DNA sequencing indicated that all patients were compound heterozygous for missense mutations in the gene for the catalytic subunit of DNA polymerase γ (POLG1). Three of the identified mutations, H1110Y, H1134R and E1136K, are novel. Fibroblasts from the patients with infantile hepatocerebral mtDNA depletion syndrome showed a progressive loss of mtDNA during culturing, whereas fibroblasts from patients with Leigh syndrome or Alpers-Huttenlocher syndrome showed reduced but stable levels of mtDNA. DNA polymerase γ activity was below the normal range in all patient cell cultures, except for one; however, this culture showed low levels of the heterodimeric enzyme on immunoblots of mitochondrial protein resolved on native gels. Levels of mtDNA and DNA polymerase γ activity were normal in parental fibroblasts consistent with the observation that all carriers were asymptomatic. The cell culture experiments established the pathogenicity of the identified POLG1 mutations and helped to define the molecular mechanisms responsible for mtDNA depletion in the patients' tissues. The assays may facilitate the identification of those patients in whom screening for POLG1 mutations would be most appropriate.

**56. Patterns of conservation in dystrophin and utrophin rod domains suggest specific functions**

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The rod domain of dystrophin comprises ~75% of the protein, and is the target of the vast majority of mutations in both DMD and BMD; indeed, most BMD mutations act by directly affecting the structure of the rod domain. To our knowledge, the only function so far attributed to the rod domain, other than that of being a "molecular ruler", is a relatively non-specific ability to bind actin; here the actin-binding capacities of dystrophin and utrophin lie in mutually exclusive regions, with binding mediated by a relatively generic property (high pI) of the interacting repeats. We used sequence acquisition and exon-informed alignment to study the pattern of conservation of residues in the rod domains of vertebrate dystrophins, utrophins, and ancestral dystrophin proteins of animals such as lampreys. We find a striking pattern of conservation which cannot be explained by the known actin-binding activity, and which therefore warrants some other explanation. Specifically, regions near the two ends of the rod domain and in spectrin repeats 8-11 are strongly conserved in ways which cannot be explained by structure alone. These regions may therefore represent sites for specific protein-protein interactions. In addition, we show that contrary to expectations, pI is poorly constrained durin evolution.

**57. Neuromuscular features of episodic ataxia: a guide to EMG investigation**

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**Background:** The episodic ataxias (EA) are rare genetic disorders of voltage gated ion channels manifesting central and peripheral nervous system features. Episodic Ataxia Type 1 is caused by mutations in the potassium channel Kv1.1 subunit which is expressed in cerebellar Purkinje cells and juxta-paranodal peripheral nerve axons. It contributes to nerve epolarisation after an action potential. Channel dysfunction results in nerve hyperexcitability. Episodic Ataxia Type 2 is caused by mutations in the Cav2.1 subunit of the voltage gated calcium channel expressed at the presynaptic neuromuscular junction. Although CNS features of EA are striking, each condition has distinctive neuromuscular manifestations. These features should be carefully defined as they can direct clinical and genetic diagnosis, and therefore treatment and prognosis.
**Aim:** To describe neuromuscular findings in genetically confirmed subjects with EA1 and EA2 and to outline an approach to EMG investigation.
**Methods:** Report of two patients and literature review neuromuscular manifestations of EA1 and EA2.
**Results:** A repetitive compound muscle action potential was seen in response to a single stimulus during motor nerve conduction studies in the subject with genetically confirmed EA1. The subject with EA2 did not show neuromuscula junction instability, although this is reported in selected subjects with EA2.
**Conclusion:** Clinical neurophysiology studies are critically important in assessment of Episodic Ataxia as they can distinguish between EA1 and EA2. EMG finding of neuromyotonia is the hallmark of EA1 whereas this is not seen in EA2.

**58. *In vivo* target gene selection by Pax7**

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Pax genes encode transcription factors that are critical regulators of key developmental processes in animals. Pax7 functions in the developing nervous system and musculature, and in adult muscle specifies myogenic satellite cells, required for regeneration. Pax7 appears to direct cell fate by influencing differentiation, proliferation, and apoptosis, however, its precise functional role has remained elusive. As a sequence-specific DNA-binding transcription factor, any direct functional role played by Pax7 is mediated through target gene selection. Thus, we have used a chromatin immunoprecipitation (ChIP) cloning assay to isolate cis-regulatory regions bound by Pax7 in mouse foetal tissue in vivo. Through sequencing and genomic localisation of a library of Pax7-bound DNA we have identified 34 target genes, with occupancy of a selectio confirmed using independent chromatin enrichment tests. We have initially assessed the capacity of Pax7 to regulate transcription from these loci through forced expression of Pax7 alternate transcripts (differing significantly in their DN binding domain) in
cultured cells, then analysed target gene expression levels using RT-PCR. We show that Pax7 directly occupies sites within genes encoding transcription factors Gbx1 and Eya4, the cytokine receptor CntfR, the potassium channel Kcnk2, and the signal transduction kinase Camk1d in vivo and regulates the transcriptional state of these genes in cultured cells. We are currently assessing the relevance of Pax7 regulation of these target genes to adult muscle regeneration. This strategy provides a powerful approach for discovery of target genes regulated by a transcription factor, and further elucidates the functional role played by Pax7.

**59. Mild POMGnT1 mutations underlie a novel limb girdle muscular dystrophy variant**

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The secondary dystroglycanopathies are a heterogeneous group of muscular dystrophies characterised by hypoglycosylation of _-dystroglycan (ADG) on skeletal muscle biopsy. To date, the identification of POMGnT1 mutations has been restricted to patients with CMD and brain abnormalities. The majority of these patients have a phenotype consistent with muscle eye brain disease (MEB), although some cases resembling Walker-Warburg Syndrome have also been reported. The objective of this study was to investigate whether mutations in this gene could be responsible for milder allelic variants of muscular dystrophy as demonstrated in other secondary dystroglycanopathy genes. A patient with a Limb Girdle Muscular Dystrophy (LGMD) phenotype, presenting at 12 years of age with severe myopia, normal intellect and reduced ADG immunolabeling in skeletal muscle was screened for mutations in the six known or putative glycosyltransferase genes. We identified homozygous POMGnT1 missense mutation (c.1666G>A, p.Asp556Asn) in this patient. No mutations were detected in the five other secondary dystroglycanopathy genes. Detailed enzymatic studies performed on the patient fibroblast's showed an altered kinetic profile, less marked than in patients with MEB, and in keeping with the patient's relatively mild phenotype. The reduction of ADG labelling was subtle on immunocytochemistry and no significant reduction in molecular weight was observed on Western blot. This highlights the difficulty in detecting mild ADG abnormalities as has been previously observed with mild FKRP mutations in LGMD2I. This report significantly widens the spectrum of disorders known to result from mutations in POMGnT1 to include LGMD without mental retardation and represents the mildest POMGnT1 deficient patient described.

**60. Clinical aspects of McArdle disease in the UK**

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Clinical data from 60 patients referred to a specialist McArdle disease clinic between 1998-2007 will be presented. Of these, 55 had previously been diagnosed by muscle biopsy, in 10 the diagnosis could not be confirmed, instead: two had Becker muscular dystrophy, one mini-core myopathy and the remainder chronic fatigue syndrome. Clinical notes were audited for the following information: age of first symptoms, age at diagnosis, symptoms, frequency of myoglobinuria, frequency of acute renal failure, creatine kinase, DNA studies for the hotspot mutations R50x and G205s. 92% of patients recalled symptoms before 20 years of age, but only 53% were diagnosed by this time. Exercise induced myalgia occurred in 95%, 76% recognised a second wind, 61% had myoglobinuria, acute renal failure occurred in 11%, 39% suffered chronic pain or fatigue and 30% had received psychiatric treatment for depression, anxiety or bipolar disorder. Muscle hypertrophy was present in 34%, and mild upper limb and truncal weakness occurred in 16% all of whom were over 40 years. A 12 minute walk test showed marked variability in exercise performance ranging from 71m-1250m. Creatine kinase was raised in all but one (mean 2,700 iu/l). DNA studies showed at least one mutated allele for R50x in 83% of patients.

**61. Model systems for developing therapies for McArdle disease**

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McArdle Disease (Glycogen Storage Disease V) is a rare inherited metabolic muscle disease caused by mutations in the Muscle Glycogen Phosphorylase (MGP) gene. MGP (myophosphorylase) is a muscle-specific enzyme needed for glycogen metabolism. The most common mutations in Caucasian patients are the early stop codon R50X (incidence around 80%) and missense G205S (incidence around 10%) mutations. Existing animal models do not have these mutations and there are no existing cell culture models of McArdle's Disease, except from patient muscle biopsies. In order to study therapeutic approaches such as correction or readthrough of the these mutations, we have made cell culture models of McArdle Disease using in-vitro site-directed mutagenesis to introduce the R50X and G205S mutations into full length wildtype mouse MGP cDNA in the mammalian expression vector pCIneo. Stable cell lines were created by transfection into CHO-K1 cells, which does not express endogenous MGP. RT-PCR has shown that MGP mRNA is being transcribed by cells transfected with mutant R50X and G205S. Gene copy number, levels of mRNA and of muscle glycogen phosphorylase expression have been quantified. MGP was detected immunologically in cells with wildtype MGP but not in cells with R50X or G205S mutant MGP. The cell models may be useful in identifying drugs or other treatments that might enable functional MGP production from the mutant form of the gene. This study was supported by Association for Glycogen Storage Disease and the RJAH Institute of Orthopaedics.

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