MYO-MRI

MRI & MRS IN NEUROMUSCULAR DISEASE

Imaging in Neuromuscular Disease 2017

First International Conference on Imaging in Neuromuscular Disease

19th – 21st November 2017 | Berlin, Germany

Programme Committee

Volker Straub Newcastle, UK	**Hermien Kan** Leiden, NL	**Pierre Carlier** Paris, FR	**Andrew Blamire** Newcastle, UK
Tarek Yousry London, UK	**Gustav Strijkers** Amsterdam, NL	**Diana Ribeiro** London, UK	**Glenn Walter** Florida, US

Platinum Sponsor

SAREPTA THERAPEUTICS

This page intentionally left blank

Abstracts from "Imaging in Neuromuscular Disease 2017: First International Conference on Imaging in Neuromuscular Disease, 19th – 21st November 2017, Berlin, Germany"

CONTENTS

Dignostic Muscle Imaging

New Imaging Techniques

Quantitative Muscle Imaging

Journal of Neuromuscular Diseases x (2017) S5–S57
IOS Press

Abstracts 1-26

Diagnostic Muscle Imaging

1

Longitudinal upper limb muscle MRI in dysferlinopathy: examining the relationship between semiquantitative MRI and physiotherapy outcome measures

Roberto Fernández-Torrón[1,2], Meredith K. James[1], Jordi Díaz-Manera[3], Anna Mayhew[1], Michele Eagle[1], Robert Muni[1], Fiona Smith[4], Andrew M. Blamire[4], Pierre G. Carlier MD[5], Helen Sutherland[1], Tanya Stojkovic[6], Maggie C. Walter[7], Eva M. Coppenrath[8], Anthony Peduto[9], Kristi J. Jones[10], Anne Marie Sawyer[11], Carolina Tesi Rocha[12], John W. Day[12], The Jain COS Consortium, Kate Bushby[1], Volker Straub[1]

[1]The John Walton Muscular Dystrophy Research Centre, Newcastle upon Tyne, UK [2]Neuromuscular Area, Biodonostia Health Research Institute, Neurology Service, Donostia University Hospital, Donostia-San Sebastian, Spain [3]Neuromuscular Disorders Unit, Neurology Department, Hospital de la Santa Creu i Sant Pau, Barcelona, Spain [4]Magnetic Resonance Centre, Institute for Cellular Medicine, Newcastle University [5]AIM & CEA NMR Laboratory, Institute of Myology, Pitié-Salpêtrière University Hospital, 47-83, Paris, France [6]Institut de Myologie, AP-HP, G.H. Pitié-Salpêtrière 47-83, Boulevard de l'Hôpital, Paris, France [7]Friedrich-Baur-Institute, Dept. of Neurology, Ludwig-Maximilians-University of Munich, Germany [8]Department of Clinical Radiology, Ludwig-Maximilians-University Munich, Germany [9]Department of Radiology, Westmead Hosptial, Faculty of Health Sciences, University of Sydney, Australia [10]Institute for Neuroscience and Muscle Research, Children's Hospital at Westmead, University of Sydney, Australia [11]Lucas Centre for Imaging, Stanford University School of Medicine, Stanford, CA, USA [12]Department of Neurology and Neurological Sciences, Stanford University School of Medicine; Stanford, CA, USA

Dysferlinopathies are caused by mutations in the DYSF gene. Patients may present with isolated hyperCKemia, limb girdle muscle weakness or predominant weakness of the lower legs. The Jain Foundation is funding The Clinical Outcome Study for Dysferlinopathy, a multi-centre natural history study in a large cohort of dysferlinopathy patients.

Previous small studies have reported an involvement of scapular muscles on muscle MRI but no longitudinal studies have been reported so far regarding upper limb involvement in muscle MRI and its relationship with physiotherapy functional outcome measures.

To describe baseline and year 3 upper limb muscle MRI involvement in a large cohort of dysferlinopathy patients. To examine the longitudinal correlation between muscle MRI semiquantitative scoring and functional outcome measures.

203 patients (11 to 86 years old) were enrolled in the JAIN COS study across 14 centers (Europe, USA, Australia, Japan). Patients underwent physiotherapy, medical and MRI assessments. 74 patients underwent upper limb muscle MRI at the time of the enrollment and 61 patients had upper limb muscle MRI at year 3. Scans were acquired from different systems and manufacturers at 1.5T and 3T. Muscles were scored on axial T1-weighted sequences with the semiquantitative Mercuri visual scale modified by Fisher. Physiotherapy assessments included muscle strength (manual muscle testing; hand held dynamometry) and functional ability evaluations (Performance of Upper limb, Brooke test). Change between baseline and year year 3 muscle MRI was assessed using Wilcoxon's Signed Rank Tests and statistical significance was set at p=0.05.

The subscapularis (80.8%), latissimus dorsi (82.6%), infraspinatus (73.8%) and supraspinatus (72.8%) were the most affected scapular muscles at baseline. The subscapularis muscle was involved in some patients without proximal upper limb symptoms or dysfunction. The biceps brachii (57.1%) and the anterior muscles of the forearm (53.8%) were the most affected muscles from the arm and forearm at baseline. The tongue (34.2%) and the cervical paraspinal muscles (24.6%) were most commonly involved at baseline. Cranial muscles and cervical did not show longitudinal changes apart for tongue and cervical paraspinal muscles. Only the brachialis, triceps, and the posterior muscles from the forearm did not show longitudinal changes. Every muscle

ISSN 2214-3599/17/$35.00

group correlated with appropiate functional outcome measures.

The subscapularis muscle and the latissimus were the most affected muscles at baseline. Only the tongue and cervical muscles showed changes at year 3 but nearly all scapular and arm muscles showed longitudinal changes. Muscle semiquantitative scoring correlated with functional outcome measures in upper limb in dysferlinopathy patients.

2

Whole-Body MR pattern in Partial laminin alpha 2 deficient patients due to LAMA2 gene mutations

Quijano-Roy S[1], Haberlova J[2], Castiglioni C[3], Gomez-Andrés D[4], Stojkovic T[5], Rivier F[6], Leboucq N[6], Fattori F[7], Bertini E[7], Dabaj I[1], Cavassa E[1], Tordjman M[8], Beroud C[7], Munell F[4], Kynčl M[2], Sanchez A[4], Ortega X[9], Carlier RY[8]

[1]Unité Neuromusculaire Pédiatrique, Service de Pédiatrie, Hôpital Universitaire R. Poincaré, Garches, France [2]Faculty Hospital, Motol, Prague, Czech Republic [3]Pediatric Neurology Department, Clinica Las Condes [4]Child Neurology Unit, Hospital Universitari Vall d'Hebron, Vall d'Hebron Institut de Recerca, Barcelona, Spain [5]Neurology Department, Pitié-Salpêtrière Hospital, Institute of Myology, Paris, France [6]Centre Maladies Neuromusculaires, Service de Pédiatrie, CHU Montpellier, France [7]Laboratory of Molecular Medicine, Bambino Jesus Hospital, Roma, Italy [8]Service d'Imagerie Médicale, Pôle Neuro-locomoteur, Hôpital R. Poincaré, Garches, France [9]Diagnostic Imaging Service. Clinica Las Condes. Santiago, Chile

Objective: Laminin alpha2 (merosin) deficient muscular dystrophy (MDC1A) is a frequent type of congenital muscular dystrophy (CMD) and it is due to LAMA2 gene mutations. Differential diagnosis may be difficult in partial deficiency patients who show also hyper-CKemia and present later than classic CMDs or with milder symptoms (dystroglycanopathies, early-onset limb girdle muscular dystrophies). A preliminary study in 8 patients with lower limb MRI (Haberlova et al 2017) showed homogeneous pattern but overlapping signs with other CMDs, in particular those involving the extracellular matrix (Collagen VI).

To confirm the homogeneity of the previously described pattern of lower limb muscle MR in partial merosin deficiency patients when using the whole body MR (WBMR).

Methods: WBMR was analyzed in 12 patients, all acquired ambulation, one had mental retardation and refractory epilepsy, four are children and eight are adults. All patients carried two causal mutations in the LAMA2 gene.

Results: Lower limb studies showed selective fatty infiltration of gluteus medius and minimum, adductor magnus, soleus and gactrocnemius muscles; relative sparing of gracilis and tibialis posterior muscles. The scanning of other parts of the body identified further similarities: selective involvement was observed in temporalis, subscapularis, latissimus dorsi, serratus anterior, paraspinal and perineal muscles. Pseudo-Collagen VI signs were less prominent and less frequent than in collagen VI myopathies. Brain white matter changes were revealed by using STIR sequences as part of the protocol.

In conclusion, whole body MR confirms previous findings of studies in partial merosin deficient patients at the lower limbs and increases the diagnostic impact of muscle scanning from head to toes. Further studies and analysis are necessary to precise the differences with other overlapping muscular dystrophies (in particular dystroglycanopathies) and retractile myopathies (collagen VI).

3

MYO-MRI ScanBank –A secure, online imaging database for neuromuscular MRI scans

Jodi Warman Chardon[1,2], Olav Veldhuizen[2], Roberto Fernandez Torron[2,3], Niels Steen Krogh[3], Volker Straub[2]

[1]The Ottawa Hospital OHRI/Children's Hospital of Eastern Ontario CHEORI, Ottawa, Canada [2]John Walton Muscular Dystrophy Research Centre, Institute of Genetic Medicine, Newcastle University, Newcastle Upon Tyne, United Kingdom [3]Neurology Department. Hospital Donostia, San Sebastián, Spain [4]ZiteLab ApS, Frederiksberg, Denmark

Muscle magnetic resonance Imaging (muscle MRI) allows for non-invasive and comprehensive assessment of striated muscles. Muscle MRI can identify selective patterns of muscle atrophy, fatty degeneration and muscle edema that help to characterize and distinguish between different genetic muscle diseases. However, in spite of the increasing clinical use, muscle MRI has not been systematically assessed for diagnostic utility in patients with genetic myopathies.

We are optimizing the MYO-MRI ScanBank, a secure, online imaging portal to view anonymized patient muscle MR images across the world.

MYO-MRI ScanBank is an imaging platform designed to assemble diagnostic muscle MRI scans that are currently scattered across neuromuscular centres internationally, to develop an inventory of images from a broad spectrum of NMD in one database. This imaging software tool is currently being optimized to integrate a rapid method of muscle grading for each MR image within the context of this study.

We have developed the MYO-MRI ScanBank image repository to 1) share characteristic whole body MR images with neuromuscular disease specialists across the world and 2) permit rapid and standardized muscle grading of these muscle images.

With the ScanBank currently being optimized for multicenter use, we will next assess the diagnostic accuracy and rate of patients with genetic myopathies based on patterns of affected muscles on MRI in a large multinational study. By collectively pooling MRI scans for patients with muscle disease, we will better identify selective patterns of muscle atrophy, fatty degeneration and muscle edema that help to distinguish between different types of genetic myopathies. Recognizing patterns of pathology by muscle imaging can help to guide genetic testing and to avoid the more invasive procedure of a muscle biopsy. Conversely, as massive parallel sequencing is now more commonly used as the initial step in diagnostic testing, imaging techniques may be useful to confirm or exclude if a variant of uncertain significance is indeed disease causing and compatible with a pattern of pathology on imaging. Finally, MRI may be useful as an imaging outcome measure in quantifying disease progression, which is critical given the recent increment in rare disease therapeutic trials.

4

Diaphragm structure-function relationship in Duchenne Muscolar Distrophy (DMD) assessed by multivolume MRI: preliminary results

F. Pennati[1,2], F. Arrigoni[2], M.G. D'Angelo[2], A. Aliverti[1]

[1]Dipartimento di Elettronica, Informazione e Bioingegneria, Politecnico di Milano [2]IRCCS E. Medea, Bosisio Parini

Diaphragm evaluation is essential in this era of developing therapeutic strategies to improve muscular, respiratory and cardiac function in Duchenne muscular dystrophy (DMD). Nevertheless, traditional techniques evaluating diaphragm function are invasive, complex and may use ionizing radiations (i.e. CT). Muscle MRI is increasingly being used in the evaluation of disease severity in neuro-muscular diseases such as DMD.

The present study was designed to investigate the feasibility of multivolume MRI, as a non-invasive imaging modality, for the assessment of the diaphragm impairment in patients with DMD. In particular, we aimed to correlate MRI measurement of diaphragm mobility and structure with age, lung volume variation and pulmonary function tests (FEV1 %predicted, forced expiratory volume in 1 second and FVC %predicted, forced vital capacity).

17 DMD patients (age 17.5±5.3 years, FEV1 59.4±28.7 %pred, FVC 60.1±28.6 %pred) were acquired on a 3T-scanner at suspended full-expiration (EXP) and suspended full-inspiration (INSP), using a multi-point gradient echo Dixon sequence. INSP and EXP lung volumes were automatically segmented by a custom-software and the surface of the diaphragm was reconstructed as the bottom surface of the left and the right lung. We measured: 1) craniocaudal diaphragmatic excursion as the median vertical distance between the EXP and INSP diaphragm surfaces; 2) muscle fatty infiltration as the amount of intramuscular adipose tissue (f/(f+w)) in regions of interest drawn manually within the diaphragmatic muscle by using the Medical Image Processing, Analysis, and Visualization (MIPAV) software. With age, the diaphragm was characterized by a decreased vertical excursion (normalized to EXP lung volume, $r=-0.47$, $p=0.05$) and an increased fatty infiltration ($r=0.86$, $p<0.001$).

Diaphragm excursion decreased with decreasing FEV1 %pred ($r=0.46$, $p=0.01$), FVC %pred (0.42, $p=0.01$) and lung volume variation ($r=0.82$, $p<0.001$).

Fatty infiltration increased with decreasing FEV1 %pred ($r=-0.69$, $p<0.001$), FVC %pred (-0.78, $p=0.01$) and lung volume variation ($r=-0.65$, $p<0.001$).

Preliminary results showed that the progressive impairment of the diaphragm, in terms of both mobility (diaphragm excursion) and structure (fatty infiltration), is highly related to age and pulmonary function tests in DMD. The results suggest that MRI might represent a new and noninvasive tool for the functional and structural assessment of the diaphragm.

5

Magnetic Resonance Image in Oculopharyngeal muscle dystrophy

Alicia Alonso-Jiménez[1,2], Aída Alejaldre-Monforte[1,2], Cristina Dominguez-González[2,3], Elena Cortes-Vicente[1,2], Ricardo Rojas-García[1,2], Giorgio Tasca[4], Robert Carlier[5], Mauro Monforte[4], Pascal Laforet[5], Gerardo Gutiérrez-Gutierrez[2,6], Adolfo López de Munain[7], Roberto Fernández-Torrón[7], Isabel Illa[1,2], Jordi Diaz-Manera[1,2]

[1]Neuromuscular Disorders Unit. Neurology Department, Universitat Autònoma de Barcelona, Hospital de la Santa Creu I Sant Pau, Barcelona, Spain [2]Centro de Investigación Biomédica en Red en Enfermedades Raras [3]Neuromuscular Disorders Unit, Neurology Department, Hospital 12 de Octubre, Madrid, Spain [4]Istituto di Neurologia, Università Cattolica del Sacro Cuore, Fondazione Policlinico Universitario "A. Gemelli", Rome, Italy [5]Centre de référence de pathologie neuromusculaire Paris-Est, institut de myologie, groupe hospitalier Pitié-Salpêtrière, Paris, France [6]Neuromuscular disorders unit, Hospital Universitario Infanta Sofía, San Sebastián de los Reyes, Madrid, Spain [7]Neuromuscular Unit, Neurology Department, Hospital Universitario Donostia, CIBERNED, Institulo Biodonosta, San Sebastián, Spain

In the past decade, muscle magnetic resonance image (MRI) has proved to be useful for diagnosis and follow-up in many muscle dystrophies.

Oculopharyngeal muscle dystrophy (OPMD) is an autosomal dominant inherited muscle dystrophy characterized by late onset of progressive ptosis, dysphagia and lower limb weakness. There are only a few articles describing the skeletal muscle findings on MRI scans in a small number of patients with OPMD.

Our aim is to describe the radiological features of a large cohort of patients with OPMD.

We are conducting an international and multicentric study with the objective of describing the pattern of fat infiltration in muscle imaging in a large cohort of patients, and correlating it with clinical function. We have analyzed axial T1-W sequences of whole-body MRIs using modified Mercuri scale to estimate the fat infiltration of each muscle. We have also collected clinical data from all the patients analyzed.

We present the results of the analysis of the first 20 patients analyzed. Our results show a distinct pattern of muscle involvement. Soleus, tongue and adductor magnus are affected in nearly all patients. Peroneus, gluteus and the posterior compartment of the thigh are also affected in most patients, whereas the gastrocnemius and the vasti are not affected until the late stages of the disease. In the trunk, anterior serratus is the most frequently infiltrated muscle. In contrast to this, upper limbs and pelvic floor muscles are spared in the majority of patients. We have found a positive correlation between clinical situation and muscle MRI changes.

In conclusion, the pattern we are describing is consistent among patients with OPMD and different from other dystrophies. Therefore, MRI could be an useful tool for the diagnosis of oculopharyngeal muscle dystrophy.

6

Muscle magnetic resonance imaging in VCP-related multi-system proteinopathy (IBMPFD): is the clue in the "fat pockets"?

Roberto Fernández-Torrón[1,2], Chiara Marini Bettolo[1], Bjarne Udd[3], Giorgio Tasca[4], Hanns Lochmüller[1], Volker Straub[1], Teresinha Evangelista[1]

[1]The John Walton Muscular Dystrophy Research Centre, MRC Centre for Neuromuscular Diseases, Institute of Genetic Medicine, Newcastle University, Newcastle upon Tyne, UK [2]Neuroscience Area, Biodonostia Health Research Institute, San Sebastián, Spain. Center for Biomedical Research in the Neurodegenerative Diseases (CIBERNED) Network, Spain. Instituto Carlos III, Ministry of Economy and Competitiveness, Madrid, Spain. [3]Neuromuscular Research Center, Tampere University Hospital and University of Tampere, Finland; Folkhälsan Institute of Genetics and the Department of Medical Genetics, Haartman Institute, University of Helsinki, Helsinki, Finland; Department of Neurology, Vaasa Central Hospital, Vaasa, Finland [4]Institute of Neurology, Policlinico "A. Gemelli" Foundation University Hospital, Rome, Italy

Hereditary inclusion body myopathy with Paget disease of the bone and frontotemporal dementia (IBMPFD), a rare autosomal dominant disorder due to mutations in VCP, has recently been named VCP-related multi-system proteinopathy. A better understanding of the natural history and muscle MRI changes is believed to be of utility for future clinical trials.

We describe clinical and muscle-MRI findings in 14 individuals with IBMPFD and a predominant myopathic phenotype.

9 males and 5 females with VCP mutations were scanned in a 1.5T MR; lower limb axial T1-weighted and STIR images were analysed. Pelvic, thigh and lower leg muscles were scored bilaterally according to the Mercuri scale on axial T1-weighted sequences.

The mean age of disease onset was 41.8 years (range 31-68 years). The most frequent phenotype was a proximo-distal myopathy. The most common mutation was p.R155H (exon 5) . The mean age at MRI was 50.13 years (range 33-77 years). The most severely affected muscles were the gastrocnemius medialis, gluteus minimus, soleus, adductor magnus and vastus intermedius. In 12 patients we observed focal areas of muscle replacement by fat ("fat pockets"). STIR abnormalities were detected in 6 patients.

Lower limb MRI in IBMPFD showed proximal and distal muscle fat replacement. Focal intramuscular areas of fat replacement might be a diagnostic marker for IBMFPD.

7

Assessment of Qualitative Magnetic Resonance Imaging in the Diagnosis of Neuromuscular Diseases: A Retrospective 1.5T Single Centre Study in Ghana

Albert D. Piersson and Philip N. Gorleku

University of Cape Coast, Department of Imaging Technology and Sonography, University Post Office, Cape Coast, Central Region, Ghana

Neuromuscular diseases are debilitating conditions which comprise of a large number of clinically and diverse disorders affecting skeletal muscle and their nerves, which can be life-threatening. Non-invasive, non-ionizing magnetic resonance imaging technique is of essential value to interrogate muscle structure, function, and metabolism in such diseases due to its inherent contrast ability and sensitivity to a wide range of soft-tissue pathologies. MRI is of tremendous importance for diagnosis, characterization of various muscle pathologies, as well as for the development and evaluation of treatment strategies.

To evaluate the utility of magnetic resonance imaging (MRI) in the diagnosis of patients who presented with neuromuscular disease.

Qualitative MRI data was obtained from a 1.5T scanner in a tertiary healthcare facility in Ghana. Fifty-seven patients (32 males; 25 females) $\geq$ 18 years of age who presented with clinical history of a form of neuromuscular disease were identified. T1W, T2W, STIR, FLAIR, and FatSat were obtained in coronal and axial planes with 5mm slice thickness and 20% interslice gap. Semi-quantitative grading (0-4 scale) was carried out in addition to subjective oedema grading (0-3 scale).

78% hyperintense T1 signal intensity (SI) and 68% hyperintense T2 SI demonstrated that fatty infiltration was the most common MRI features presented on gluteus maximus, calf, and pelvic muscles. There was a significant correlation between the mean T2 values and semi-quantitative MR imaging grade of both fatty infiltration and oedema.

MRI is a non-invasive, effective method for diagnosis, monitoring treatment and future follow-up of neuromuscular diseases. Qualitative and semi-qualitative grading provides robust methods to evaluate neuromuscular diseases using MRI. Fatty infiltration and oedema are prevalent features seen on MRI images of patients with neuromuscular diseases.

8

Pattern recognition in neuromuscular children population. Experience of Russian Children Neuromuscular Center

Vlodavets D.V., Kazakov D.O.

Russian Children Neuromuscular Center, Veltischev Clinical Pediatric Research Institute of Pirogov Russian National Research Medical University, Moscow, Russia.

Muscle imaging diagnosis in neuromuscular field is based on patterns muscle involvement. In Russian Children Neuromuscular Center we use MRI scanner model Toshiba Vantage Excelart with field strength 1,5T. Protocol included T1w and T2w sequences with slice thickness 5 mm and gaps 10 mm. We use visual grading of fat infiltration on T1w spin echo images and Lamminen-Mercuri scale (in modification 2002) for evaluation which is rather satisfactory for patters recognition purposes. In diagnostic imaging we use fat-saturated T2w sequences (STIR) to detect inflammatory myopathies. Unfortunately we are cannot do multi-echo pulse sequence to measure fat fraction and DIXON sequences because the

limits of our scanner. We analyzed scans of 816 patients that were investigated for the period of last 4 years with suspected neuromuscular disorder. Typically there were scanned thighs and ankles of the patients.

Structure of the examined patients is listed below. 163 patients had no any muscle involvement that could be seen on T1w and T2w sequences of MRI images. 180 had some non-specific degenerative changes in some muscles but it was impossible to use this for patters recognition analysis. Other 473 patients had very prominent patters of muscle involvement of different neuromuscular disorders. The greatest group had 201 DMD patients, and 15 BMD patients. On the 2-nd place there were 61 patients with collagenopathies. On the 3-rd place there were 45 patients with different types of SMA. Other disorders that was detected in our collection: congenital myopathy (RYR1) – 21 patients, LGMD 2A (CAPN3) – 16 patients, Emery-Dreifuss muscular dystrophy – 15 patients, congenital muscular dystrophy type 1A (LAMA2) – 15 patients, FSHD – 13 patients, polymyositis – 12 patients, CMT (different types) – 12 patients, LGMD 2D (SGCA) – 9 patients, LGMD 2B (DYSF) – 8 patients, LGMD 2L (ANO5)– 8 patients, IBM – 6 patients, congenital nemaline myopathy (ACTA1, NEB) – 5 patients, LGMD 2I (FKRP) – 5 patients, myotonia congenita (CLCN1) – 2 patients, Pompe disease (GAA) – 2 patients, myotubular myopathy (MYOT) – 1 patient, myofibrillar myopathy (DES) – 1 patient.

T1w and T2w sequences could be used for patters recognition purposes in patients with neuromuscular disorders. Muscle MRI could provide confirmation of the diagnosis and gives additional information about the disease progression. Most impressive is that muscle MRI is noninvasive technique compared with muscle biopsy or needle EMG study.

9

Longitudinal evaluation of muscle involvement in FSHD by muscle MRI

Mauro Monforte[1], Giorgio Tasca[1], Pierfrancesco Ottaviani[2], Francesco Laschena[2], Enzo Ricci[1]

[1]Istituto di Neurologia, Università Cattolica del Sacro Cuore, Fondazione Policlinico Universitario "A. Gemelli", Rome, Italy [2]Dipartimento di Radiologia, IDI IRCCS Rome, Italy

Cross sectional studies reported the most frequent muscle imaging patterns in Facioscapulohumeral muscular dystrophy (FSHD), whilst only limited longitudinal data are available at the moment. With many clinical trials in progress or at the horizon, the knowledge of the natural history of FSHD is of paramount importance to understand the impact of new therapies. Given the peculiar muscle-by-muscle involvement in FSHD, muscle MRI represents an invaluable tool to explore the modalities and rate of disease progression.

To assess yearly disease progression in FSHD patients at single muscle level using muscle MRI.

A cohort of 70 genetically confirmed FSHD patients underwent a baseline and a 1 year follow-up lower limb muscle MRI. T1 weighted (T1w) and T2 weighted-short tau inversion recovery (T2w-STIR) sequences were used to assess the presence of fatty replacement (using a semiquantitative visual score) and of oedema (using a binary score, i.e presence/absence). The scans were also evaluated by unblinded direct comparison to detect the changes not captured by the previous scoring system, and the possible changes detected by this evaluation were classified into 16 different categories.

We report: 1) the percentage of muscles that changed their score of fat infiltration; 2) the percentage of muscles that, although not showing a change in their score, demonstrated clear signs of progression in the direct unblinded comparison; 2) the rate of appearance/disappearance of T2w-STIR+ lesions; 3) the relation existing between T2w-STIR+ lesions and progression of fat changes in each muscle. Results are still under evaluation and will be presented at the congress.

Data derived from this longitudinal MRI study contribute to delineate the natural history of FSHD, thus providing useful evidence for future clinical trials in this disease.

10

Whole Body MRI of a cohort of Egyptian patients with Limb-girdle muscular dystrophy

Fahmy, N; Ashour, S; Ihab, S; Abu El-Fetouh, K; Saadawy, A

Faculty of Medicne, Ain Shams University

Limb-girdle muscular dystrophy (LGMD) is a heterogonous group of muscle diseases with several genetic mutations that cause diagnostic difficulty. Each subtype of LGMD has specific muscles affected that give the clinical pattern of this subtype. Muscle MRI has become a useful tool for diagnosis of patients with limb girdle muscle dystrophies (LGMD). Muscle MRI provides information on different aspects of muscle structure and function of muscles of the body. The sequences available allow to identify fatty infiltration or edema of muscles. But MRI muscles has a specific benefit in determining the individual muscles affected especially the deep muscles, thus helping greatly in determining the LGMD subtype and directing easily for the molecular diagnosis of patients.

We aimed from this work to diagnose patients with LGMD, determining its subtype and comparing the results with the molecular diagnosis of patients.

This work is a part of current work in Muscle and Nerve Research unit, Neurology department and MRI unit, Radiodiagnosis department, Faculty of Medicine, Ain Shams University. We established for the first time a protocol of Whole body MRI (WBMRI) to study patients with Nerve and Muscle disorders.

Twenty-one patients with LGMD are presented here. Clinical assessment, Laboratory study, WBMRI, Muscle Biopsy and Genetic study. WBMRI was done using 1.5 Tesla, siemens machine. T1, T2, STIR sequences of axial and coronal cuts were done.

WBMRI could reach a definite diagnosis in eleven patients, had a probable differential diagnosis in 5 patients and couldn't reach a diagnosis in another 5 patients. Patients with dysferlinopathy, Calpainopathy and Sarcoglycanopathy were very common in our small cohort and we are expecting increase of number of these patients through out our study. WBMRI easily diagnosed these subtypes of muscular dystrophy by their specific patterns.

WBMRI is a very useful tool in the diagnosis of patients with LGMD and should be used as an essential tool in any protocol of diagnosis of progressive muscular dystrophy.

11

Pattern of skeletal muscle involvement in distal myopathies

[1]Hyung Jun Park, [2]Young-Chul Choi
[1]Department of Neurology, Ewha Womans University Mokdong Hospital, Seoul, Korea [2]Department of Neurology, Yonsei University College of Medicine, Seoul, Korea

Distal myopathy is a heterogeneous group of genetic muscle diseases characterized by predominant distal muscle weakness. Distal myopathy caused by DYSF, GNE, and ADSSL1 mutations are the most common distal myopathy in Korea. Limb Muscle has recently proven to be useful for the diagnostic and prognostic marker in inherited muscular disorders.

We aimed to investigate the usefulness of lower-limb MRI in Korean patients with distal myopathy.

We retrospectively analyzed muscle MRI scans of 20 Korean patients with distal myopathy. They consisted of eleven patients with dysferlinopathy, six patients with ADSSL1 myopathy, and four patients with GNE myopathy. MRI was undertaken by a 1.5T system (Siemens Vision, Erlangen, Germany).

Eleven Korean patients with DYSF mutations demonstrated the similar fatty replacement pattern. In the thigh, muscle pathology generally seems to start in the adductor magnus muscle and then affects the semimembranosus and the vastus lateralis muscles. However, the rectus femoris, gracilis, and sartorius muscles were relatively spared. In calf muscles, the posterior compartment seemed to be the earliest and most severely affected. The STIR signal was abnormal in the adductor magnus and the medial gastrocnemius muscle. Six patients with ADSSL1 myopathy demonstrated showed more fatty replacements in the distal limb muscles than in the proximal limb and axial muscles. The sequential pattern of muscle degeneration was correlated with disease severity. At the thigh levels, predominant fatty replacements of vastus lateralis muscles were observed at the early stage. More advanced stages showed a progressive degeneration of anterior and medial compartments of thigh muscles. At the calf level, fatty replacements started at gastrocnemius muscles and followed by whole compartments of calf muscle. Four patients with GNE myopathy demonstrated the predominant fatty replacement of the adductor longus, semitendinosus and biceps

femoris short head muscles at the thigh. However, the vastus lateralis was the only muscle spared in advanced stages, while the rectus femoris, vastus intermedius and medialis showed variable signs of fatty replacement. At the calf level, tibialis anterior and extensor digitorum longus muscles were mainly affected and then the soleus, peroneus longus and tibialis posterior muscles followed.

Our result revealed the usefulness of MRI scans for pattern recognition and differential diagnosis in distal myopathy.

12

Magnetic resonance imaging analysis of micro-dystrophin treated mdx4cv muscles

Jeffrey S. Chamberlain, Joshua Park, Ravneet Vohra, Thomas Klussmann, Katrin Hollinger, Niclas Bengtsson and Donghoon Lee

The Departments of Neurology, Radiology, Biochemistry and Medicine, and the Senator Paul D. Wellstone Muscular Dystrophy Cooperative Research Center, University of Washington, Seattle, Washington, 98195 USA

We have been exploring gene therapies for DMD using AAV-mediated delivery of micro-dystrophin genes. One approach to monitoring the effects of therapy in a non-invasive manner is to use MRI methods.

The primary goal is to evaluate the phenotypic effects of AAV-mediated gene transfer of micro-dystrophin genes regulated by muscle-specific gene regulatory elements. Treated animals are monitored to assess pathophysiology, and also at various intervals by MRI.

Several unique micro-dystrophin cDNAs were generated by recombinant PCR and tested by AAV6-mediated delivery in mdx4cv mice for stability and improvement of muscle specific force generation and histology. We have also been characterizing treated and control (ctrl) mdx4cv and wild type mice using multi-parametric Magnetic Resonance Imaging (mp-MRI).

Initial studies involved imaging at 10 weeks of age for pre-treatment, then again post-treatment at 8, 16, and 24 week time points. Micro-dystrophin delivery restored numerous aspects of normal muscle histology and pathology such as decreased necrosis and resistance to contraction-induced injury. T2 relaxation values showed percentage decreases across all muscle types measured. The diffusion measurements showed a wider range of percentage changes and less statistical significance while the magnetization transfer effect measurements showed minimal change. MR images displayed hyper-intense regions of muscle that correlated with muscle pathology in histological sections. To further develop the approach we have imaged wild-type and mdx4cv groups beginning at 3 weeks of age and continuing every two weeks until 11 weeks using a 14 Tesla Avance MR spectrometer. The high-resolution MRI protocol included T1 and T2 relaxation times, magnetization transfer ratio and diffusion tensor imaging. Limb muscle T2 was significantly higher in mdx4cv compared to ctrl muscle at nearly all time points. Additionally, fractional anisotropy (FA) of all muscles was significantly different between mdx-4cv and ctrl groups. Future studies will include further analysis of AAV treated groups.

The results demonstrated quantifiable differences between the mdx4cv and normal groups. Similar MR protocols could be explored and utilized as a noninvasive means of tracking disease progression and treatment response in future clinical trials of muscular dystrophy treatment.

13

MRI quantifies lumbosacral nerve root and sciatic nerve hypertrophy in chronic inflammatory demyelinating polyradiculoneuropathy

Shah S[1], Morrow JM[2], Sinclair CDJ[1,2], Evans MRB[2], Hanna MG[2], Lunn MP[2], Reilly MM[2], Thornton JS[1,2], Yousry TA[1,2]

[1]Neuroradiological Academic Unit, UCL Institute of Neurology, London WC1N 3BG, UK [2]MRC Centre for Neuromuscular Diseases, UCL Institute of Neurology, London WC1N 3BG, UK

Chronic inflammatory demyelinating polyradiculoneuropathy (CIDP) is a treatable, immune-mediated condition characterised by progressive or relapsing motor and sensory deficits in all four limbs. The diagnosis is based on a combination of clinical, neurophysiological and supportive criteria, but can often be challenging. Consensus guidelines include hypertrophy of lumbosacral nerve roots on

MRI as a supportive criterion, although the evidence for this has not been fully validated.

To quantify the diameter and cross-sectional area of the lumbosacral nerve roots, and to explore the quantitative and qualitative imaging characteristics of the sciatic nerves, in patients with CIDP versus healthy controls using MRI.

MRI of the lumbosacral plexus and both thighs was performed at 3T in 10 patients with CIDP (7M/3F, age 52 ± 15y) and 10 healthy controls (7M/3F, age 50 ± 11y). A 3D T1-weighted gradient echo sequence was used for evaluation of the lumbosacral nerve roots, with 3D proton density (PD)-weighted, short tau inversion recovery (STIR), and dual echo T2-weighted sequences used to assess the sciatic nerves. Orthogonal diameter and cross-sectional area of the lumbosacral nerve roots were measured at three defined points. T2 values were measured along the length of each sciatic nerve by defining enclosing regions of interest on the T2w sequence, with the same method used to measure cross-sectional area at the mid-thigh level on the PDw sequence. The nerve appearance was also evaluated qualitatively on the STIR images. All assessments were performed by an observer blinded to the diagnosis.

Median diameter of the lumbosacral nerve roots was significantly increased in patients with CIDP compared to controls (CIDP 6.7mm; controls 4.8mm; p=0.0016), with a corresponding increase also shown in median cross-sectional area. The degree of nerve root hypertrophy within the CIDP group was more marked at the L5 and S1 levels, and particularly the extraforaminal portion of the L5 nerve roots (CIDP 7.7mm; controls 4.1mm). A threshold upper nerve root diameter of 6.5mm at this location distinguished between patients and controls with a sensitivity of 88% and a specificity of 89%.

Median cross-sectional area of the sciatic nerves was also increased in patients with CIDP compared to controls (CIDP 45.5sqmm; controls 29.8sqmm; p=0.008). Although median T2 values were also slightly longer in the CIDP group (57.5ms) compared to controls (54ms), this difference was not significant. Sciatic nerve STIR signal was rated as markedly hyperintense in five CIDP patients, mildly hyperintense in one, and isointense in three. In comparison, no controls were rated as markedly hyperintense, with three rated as mildly hyperintense and six as isointense.

MRI reveals significant hypertrophy of the lumbosacral nerve roots and sciatic nerves in patients with CIDP compared to controls. This study provides supportive evidence for the inclusion of lumbosacral nerve root and sciatic nerve hypertrophy on MRI in the diagnostic criteria for CIDP.

14

Proposal for a future project and working group: Muscle imaging detection of early signs in early onset myopathies

Hospital Universitari Vall d'Hebron group, Raymond Poincaré Hospital group, Universidade da Beira Interior group, Clínica Las Condes and Hospital Clínico Universidad de Chile group, Basel Children's Hospital group and National Institute of Neurological Disorders and Stroke (NINDS) group

Hospital Universitari Vall d'Hebron group (Barcelona, Spain): David Gomez Andres (Child Neurologist), Angel Sanchez-Montáñez (radiologist), Laura Costa (Child Neurologist), and Francina Munell (Child Neurologist).

Raymond Poincaré Hospital group (Garches, University of Versailles UVSQ, France): Susana Quijano-Roy (Child Neurologist, co-leader WG1 at COST action Myo-MRI), Benjamin Doré (Radiologist), Yerko Ivanovic (adult neurologist involved in arthrogryposis project) and Robert Carlier (Radiologist, member of Myo-MRI).

Universidade da Beira Interior group (Porto, Portugal): Rafael Rodrigues (Bio-engineer expert in texture analysis in muscular disorders, Project STSM COST-action) and Antonio Pinheiro (head of the laboratory expert in texture analysis).

Clínica Las Condes (1), and Hospital Clínico Universidad de Chile (2) group (Santiago de Chile, Chile): Claudia Castiglioni (Child neurologist) (1), Ximena Ortega (Radiologist) (1); Gonzalo Rojas (Informatic engineer, expert in imaging processing for the radiology department) (1); Jorge A Bevilacqua (Neurologist and Neurobiologist) (2)

Basel Children's Hospital group (Basel, Switzerland): Dirk Fischer (Child & adult neurologist and expert in muscle imaging)

National Institute of Neurological Disorders and Stroke (NINDS) group (Bethesda, MD, USA): Carsten Bönnemann (Child Neurologist and Geneticist, expert in molecular genetics and cell biology of early onset myopathies), Reghan Foley (Child Neurologist) Sarah Neuhaus, Dimah Sadee (Clinical Neuromuscular

Research Fellows at NIH), Pomi Yun (Premed Research Student with focus on imaging) and Glenn Walters (MRI Physicist with focus on neuromuscular disease at the University of Florida, collaborator on all NIH CMD imaging protocols)

Acknowledgements: COST action BM1304 (http://myo-mri.eu)

Even in the NGS era, establishing the molecular diagnosis of early onset muscle diseases (congenital myopathies (CM) and congenital muscle dystrophies (CMD)) is complicated by remarkable phenotypic heterogeneity and size and complexity of the causative genes. Hence, a diagnostic hypothesis for pre- and post- genetic testing validation continues to be of great importance.

Although histologic features facilitate diagnosis in particular cases, alternative diagnostic markers are needed to identify the most plausible candidate genes at early stages of the disease. Indeed, in-depth scrutiny of the sequence of the candidate gene(s), including intronic or regulatory regions, may payoff over other strategies of NGS data analysis which rely mostly on bioinformatic criteria for variant prioritization.

Muscle imaging is a useful tool for guiding genetic diagnosis in muscle disorders. This can be achieved using MRI as well as muscle ultrasound. Beyond the establishment of the presence of a disease of muscle, characteristic patterns of muscle involvement have been associated with specific CMD and CM forms, although most of these patterns are based on study of cases displaying the full-blown clinical pictures. Based on our preliminary data we suspect that specific imaging features are recognizable also in the early stages of many CM and CMD phenotypes. If this is the case, a systematic imaging approach to the baby/toddler with these disorders may result in an earlier diagnosis and more accurate prognosis and appropriate personalized care. We also have preliminary data to suggest different sensitivities for muscle MRI and muscle ultrasound. Ultrasound is eminently suitable to the young age as no sedation is needed and is sensitive to early myopathic findings, such as pure fiber atrophy. MRI on the other hand is superior in anatomical resolution but only sensitive the chemical changes in the composition of the muscle, most importantly driven by changes in fat and water content. The hypothesis arising from the preliminary observations is that muscle ultrasound is more sensitive to early myopathic changes but reaches maximal saturation in progressed disease early, whereas MRI is still capable of pattern differentiation in more advanced disease involving fatty replacement of muscle tissue.

We seek to establish an international collaboration to develop a project for assessing the diagnostic value of muscle imaging in children with early onset muscle disease, with the aim of identifying early specific signs of specific genetic conditions using comparative muscle imaging technologies (MRI and ultrasound).

Patients with proven molecular diagnosis of CM or CMD, with clinical onset earlier than 3 years of age and muscle imaging obtained before 6 years of age.

Phase 1. Development of multicenter MRI and ultrasound collections with phenotypic characterization.

Phase 2. Scoring of MRI and ultrasound collections, correlation with histological and genetic findings.

Phase 3. Analysis of results with heatmaps and machine learning approaches. Compare the sensitivity of MRI versus muscle ultrasound in specific diseases.

Phase 4. Texture analysis of muscle MRI and US collection in order to identify diagnosis markers in specific diseases.

Phase 5. Establishment of an international consensus of quantitative imaging based on the findings of non-quantitative imaging.

This is a proposal for a future project.

Our initiative, that have arisen as a result of the knowledge and relashionships established within the framework of the COST action BM1304, aims to develop a collaborative effort among clinicians, radiologists, geneticists, informatic engineers and physicists from different countries, to address the diagnostic value of muscle imaging in early stages of early onset muscle diseases.

15

Inventory of Phenotypic Traits in Early onset Muscle diseases (IPTEM): towards a better phenotypic annotation in muscle MRI databases of congenital muscle dystrophies and congenital myopathies

Marta Gómez García de la Banda[1], David Gómez-Andrés[1], Ángel Sanchez-Montáñez[1], Ivana Dabaj[2], Audrey Bénezit[2], Yerko Pétar Ivánovic Barbeito[2], Alfons Macaya[1], Robert Y. Carlier[2], Francina Munell[1], Susana Quijano-Roy[2]

[1]Hospital Universitari Vall d'Hebron, Barcelona, Spain [2]Raymond Poincaré Hospital group, Garches, University of Versailles UVSQ, France

In spite of the recent advances in genetics, molecular diagnosis of early onset muscle diseases is still a challenge. Whole Exome (WES) or Genome (WGS) sequencing technologies provide a huge amount of meaningless variants that make interpretation difficult and sometimes lead to erroneous diagnosis. On the other hand, some variants are difficult to identify (e.g. deep intronic mutations) or to interpret (e.g. missense variants with uncertain significance in large genes as TTN). In other situations, phenotypic traits are clear enough to use cheaper and more targeted approaches. We believe that the combination of standardized and systematic clinical and muscle MRI data could help us to improve the interpretation of WES/GES data or to select targeted strategies as first option. Although we have developed methods to systematically assess muscle MRI, there is a lack of consensus about how patients with Congenital Muscular Dystrophies (CMD) and Congenital Myopathies (CM) should be clinically described and analysed and current approaches are inaccurate.

To create an inventory of clinical signs that allows a systematic clinical characterization of patients with CMD and CM. This inventory will provide critical additional information in the evaluation of muscle MRI performance in clinical settings.

1) Extraction and re-ordering of the current HPO (Human Phenotype Ontology) terms that are annotated to each disease in the group of interest. Definition of synonyms and antonyms inside these terms.
2) Creation of a database-specific inventory with clinical signs based on HPO and clinical expertise.

We have extracted and reordered the HPO terms and created an inventory of signs based on HPO and our clinical expertise (some signs that are important in the differential diagnosis were lacking). We are now applying this inventory to our cohorts in two different centres.

It is possible to develop an inventory of clinical signs based on HPO terms that allows the systematic collection of clinical data to enrich our muscle MRI databases. We need further studies to prove its usefulness with and without muscle MRI data in the diagnosis process.

16

Whole Body MRI to differentiate inherited myopathies presenting with Rigid Spine Syndrome

Tordjman M[1], Dabaj I[2], Mompoint D[1], Laforet P[3], Felter A[1], Biyoukar M[1], Richard P[4], Quijano Roy S[2], Carlier R-Y[1] (presenting author)

[1]APHP, Medical lmaging department, pôle neuro-locomoteur, Hopital Raymond Poincaré, Groupe des hôpitaux Universitaires Paris Ile de France Ouest, 92380 Garches, France [2]APHP, Medical Pediatric department, pôle neuro-locomoteur, Hopital Raymond Poincaré, Groupe des hôpitaux Universitaires Paris Ile de France Ouest, 92380 Garches, France [3]APHP, Neurology department: Unité clinique de Pathologie Neuromusculaire; Institut de myologie; CHU Paris-GH La Pitié Salpêtrière-Charles Foix - Hôpital Pitié-Salpêtrière, France [3]APHP, Department of myogenetics; Centre de génétique moléculaire et chromosomique; CHU Paris-GH La Pitié Salpêtrière-Charles Foix - Hôpital Pitié-Salpêtrière, France

Several phenotypes are described for inherited myopathies including Rigid Spine Syndrome (RSS), which is used to designate patients affected by inherited myopathy with early spinal contractures as a prominent feature.

To elaborate an algorithm using Whole Body Muscle MRI as a tool to differentiate inherited myopathies presenting with Rigid Spine Syndrome (RSS).

73 patients from references centers in France, Brazil and Chile were included from February 2005 to December 2015 in this retrospective study. They all presented myopathies known for giving RSS (in the literature and our experience), confirmed genetically. Fatty muscle replacement was evaluated on Whole Body MRI with Mercuri scale (1 for no

infiltration and 4 for complete muscle deterioration) and with the percentage of preserved muscle (%PM) for statistical comparison between myopathies.

20 patients were affected by Pompe Disease, 8 patients had a mutation of RYR1, 17 patients had Collagen VI Myopathy, 7 patients had LMNA mutation, 8 patients had SEPN1 mutation, 5 patients had LAMA2 mutation, and 8 patients had MYH7 mutation. Each myopathy had a specific pattern of affected muscles. For example, severe infiltration of tongue was more likely found in Pompe disease (Mercuri 3; %PM=0.42; $p<0.001$) while sternocleidomastoid muscle was affected in SEPN1 mutation (Mercuri 4; %PM=0.15; $p=0.001$), as well as soleus muscle in RYR1 mutation (Mercuri 4; %PM=0.20; $p=0.01$). Patterns of affected muscles for those myopathies associated with certains aspects (like Collagene sign) allowed us to elaborate a specific algorithm for patients with RSS.

Inherited myopathies with RSS are rare and the diagnosis is often challenging. Evaluation of muscle infiltration on Whole Body MRI shows specifics patterns useful to orientate diagnosis.

17

Early involvement of the supinator muscle in Duchenne muscular dystrophy

Tartaglione T[1], Brogna C[2], Cristiano L[2], Verdolotti T[1], Pane M[2], Ficociello L[1], Fanelli L[2], Colosimo C[1], Mercuri E[2].

[1]Department of Radiology and [2]Pediatric Neurology and Nemo Clinical Centre, Università Cattolica Sacro Cuore, Fondazione A. Gemelli, Rome, Italy

In the last few years there has been increasing attention to the involvement of the upper limbs in Duchenne muscular dystrophy (DMD). Both clinical and MRI studies have shown that the first signs of upper limb weakness are related to shoulder muscles. Arm and forearm muscles are subsequently progressively affected in older boys, generally after loss of ambulation.

We report four cases of ambulant DMD boys in order to establish any early correlation with upper limb functions and MRI findings.

All 4 patients of 15, 11, 8 and 5 years, performed MRI T1 sequences at shoulder, arm and forearm level and upper limb assessment using the performance upper limb (PUL).

In the first three patients we observed a selective abnormal signal in the supinator muscle at the forearm level. All three had only minimal involvement of the shoulder muscles and no involvement of the arm muscles.

In all three the MRI abnormalities were associated with inability to perform a full supination of the forearm with less than 75% of the predicted range of movement while, using the performance of upper limb (PUL) test, they had full scores at shoulder arm and forearm level.

In the youngest patient, age 5 years, there was also a selective involvement of the supinator muscle, even if this was milder and not associated with clinical impairment.

Our results suggest that despite the well known proximal to distal gradient observed in DMD, early involvement of the supinator muscle can be found even in ambulant boys in whom more proximal muscles are completely or relatively preserved.

As the more obvious MRI changes were associated with early clinical signs of restriction of the range of supination, our findings suggest that this should be systematically investigated even in young ambulant boys in order to establish early therapeutic intervention. Further studies in larger cohorts will help to establish the consistency of this finding at different ages.

18

Patterns of upper limb muscle involvement in DMD: correlation with functional abilities

Tartaglione T[1], Brogna C[2], Cristiano L[2], Verdolotti T[1], Pane M[2], Ficociello L[1], Fanelli L[2], Colosimo C[1], Carlier P[3], Mercuri E[2].

[1]Department of Radiology and [2]Pediatric Neurology and Nemo Clinical Centre, Università Cattolica Sacro Cuore, Fondazione A. Gemelli, Rome, Italy [3]Institute de Myologie, Pitie Salpetriere, Paris

Increasing attention has been devoted to upper limb involvement in ambulant and non ambulant boys affected by Duchenne muscular dystrophy (DMD). A few recent studies have reported how clinical and MRI tools can detect early abnormalities. Most of the MRI studies have focused on the forearm.

The aim of this study was to perform a systematic assessment of shoulder, elbow and distal muscles

and to correlate MRI findings with the results of the Performance of upper limb (PUL) test that also provides information on the three upper limb segments. More specifically we wished to establish if there was any correlation between MRI and clinical findings and if MRI can identify early markers of clinical impairment.

Eighteen DMD boys, 11 ambulant and 7 non ambulant , of age between 5 and 21 years, underwent a muscle MRI assessment including T1 transverse and Dixon sequences.

All patients were also assessed using the PUL that provides a total score and subscores related to shoulder, elbow and distal dimensions.

The preliminary analysis of the results show that MRI can identify early signs of muscle involvement. Further analysis is in progress to establish a more detailed quantification of the extent of muscle impairment.

Muscle MRI can help to identify early upper limb involvement. Its use may help in the design of clinical trials in non ambulant patients.

19

The diagnostic value of MRI pattern recognition in distal myopathies

Bugiardini E[1], Morrow JM[1], Shah S[2], Wood CL[3], Lynch D[4], Pitmann A[4], Reilly MM[1], Houlden H[4], Matthews E[1], Parton M[1], Hanna MG[1], Straub V[3], Yousry TA[1,2]

[1]MRC Centre for Neuromuscular Diseases, UCL Institute of Neurology and National Hospital for Neurology and Neurosurgery, Queen Square, London, UK [2]Neuroradiological Academic Unit, UCL Institute of Neurology, & Lysholm Department of Neuroradiology, London, UK [3]John Walton Muscular Dystrophy Research Centre, Institute of Genetic Medicine, Newcastle upon Tyne, UK [4]Department of Molecular Neuroscience, UCL Institute of Neurology, Queen Square, London, UK

Distal myopathies are a diagnostically challenging group of diseases. Several publications have described a specific pattern of muscle involvement in specific genetic subgroups, but no studies have evaluated the usefulness of MRI pattern recognition in clinical practice.

We aimed to understand the value of MRI in the current clinical setting and explore its potential of optimising its clinical application.

We retrospectively audited the diagnostic workup in a distal myopathy patient cohort, reassessing the diagnosis, whilst documenting the usage of MR. We established a literature based distal myopathies MRI pattern template and assessed its diagnostic utility in terms of sensitivity, specificity and potential impact on the diagnostic workup.

Fifty-five patients were included and 38 were audited. The median time from symptoms onset to diagnosis was 12.1 years. The initial diagnostic rate was 39%, with 18% misdiagnosed as neuropathies and 13% as inclusion body myositis (IBM). The number of genes suggested by the MRI pattern analysis was significantly smaller (31) than that suggested by the clinical work up (210). MRI analysis was able to ruling out IBM in all cases. Based on 21 publications we established a MRI pattern template. Its overall sensitivity (50%) and specificity (32%) were low; its specificity for single diseases however (MYOT, MYH7, GNE, TTN-HMERF) was high (90-100%).

In the diagnostic work-up of distal myopathies, MRI is useful in guiding genetic testing and avoiding misdiagnosis (IBM). Despite its shortcomings, the literature based MRI template provides valuable support in daily clinical work. Although the overall low sensitivity and specificity limits a generalised use, it is expected that in the context of next generation sequencing, MRI's high individual disease specificity will make it central to the clinical work-up.

20

Areas of uncertainty in muscle MRI for the diagnosis of congenital muscle dystrophies and congenital myopathies: an open path for improvement

David Gómez-Andrés[1], Francina Munell[1], Ángel Sánchez-Montáñez[1], Robert Carlier[2] and Susana Quijano-Roy[2] on behalf of Pediatric disorders Statement Paper Commitee of WG1 "Improve diagnosis and understanding of muscle pathology"

[1]Hospital Universitari Vall d'Hebron. Barcelona [2]Raymond Poincaré Hospital group. Garches. University of Versailles UVSQ

Muscle MRI is an extraordinary technique to facilitate diagnosis in early onset muscle disorders (EO-MD) such as congenital muscle dystrophies (CMD) and congenital myopathies (CM). In the recent years, significant improvements in the description of the different muscle patterns in the different genetic aetiologies have been performed. However,

there are also limitations that are important to detect and adress in the future years.

To review the existing published data about muscle MRI in early onset muscle disorders in order to detect areas of uncertainty and to propose areas for future research.

We performed a systematic literature search in Pubmed to detect studies or cases that describe patterns of muscle involvement in patients with early onset (less than 5 years) and with confirmed genetic diagnosis of LAMA2-CMD, COL6-MD, COL12-MD, SEPN1-MD, FHL1-MD, integrin-related CMD, LMNA-CMD, alpha-dystrogycanopathy, CHKB-CMD, TRAPPC11-CMD, GOLGA2-CMD, TRIP4-CMD, nemaline myopathy, RYR1-related muscle disorders, MYH7-MD, myotubular CM, centronuclear CM, titinopathies, MYH2-MD, MYBPC3 CM, CNTN1 CM, sarcotubular CM, PTPLA CM and CM with opthalmoplegia related to CACNA1S.

For those EO-MD that are more common, there few case series. Moreover, patient number is usually small (<15 patients) and there is a paucity of multicentre studies. For those EO-MD that are ultra-rare, there only case reports which limits our possibilities to define a pattern.

MRI was performed in adulthood in the majority of cases. Only a few number of studies include whole-body MRI. Fewer studies include systematic reporting of muscle involvement. There is only one single study that uses quantitative techniques. Longitudinal studies are sparse.

There is still a need to investigate in muscle MRI in EO-MD. We will need international collaborative studies that use whole-body MRI, systematic reporting of muscle involvement and quantitative techniques. Longitudinal evaluation will be also an important area of research.

21

Whole-body Muscle MRI in GMPPB-related Congenital Myasthenic Syndrome

Daniel Natera-de Benito[1]; Emilio J. Inajeros Clemente[2]; Carlos Ortez[1]; María Eugenia Yoldi[3]; Anna Lia Frongia[1]; Lidia González Quereda[4]; Susana Quijano-Roy[5]; Andres Nascimento[1]

[1]Unit of Neuromuscular Disorders, Hospital Sant Joan de Déu, Barcelona (Spain) [2]Department of Radiology, Hospital Sant Joan de Déu, Barcelona (Spain) [3]Department of Pediatric Neurology, Hospital Universitario Virgen del Camino, Pamplona (Spain) [4]Genetics Department, Hospital de la Santa Creu i Sant Pau, U705 CIBERER, Barcelona (Spain) [5]AP-HP, Hôpitaux Universitaires Paris-Ile-de-France Ouest, Pôle pédiatrique, Centre de Référence Maladies Neuromusculaires (GNMH), Hôpital Raymond Poincaré , Garches; Université Versailles Saint-Quentin UVSQ (France)

Congenital myasthenic syndromes (CMS) are disorders caused by mutations in genes encoding proteins hat are essential for maintaining the integrity of neuromuscular transmission. All CMS share the clinical features of fatigable weakness. GMPPB is a recently reported CMS-associated gene involves in both the N-glycosylation and the O-mannosylation pathways.

The purpose of this study is to describe a pattern of muscle involvement in a series of three siblings who have a congenital myasthenic syndrome due to mutations in GMPPB.

We followed-up three siblings (2 males, 1 female) with a mean age of 11.7 year-old (range 7–16). All patients carried in homozygosis the previously published mutation GMPPB c.553C>T (p.Arg185Cys).

All patients had a predominantly limb-girdle pattern of muscle weakness, with moderate to severe impairment and marked waddling gait in some cases.

Patients underwent WB MRI on a 1.5-T MR imaging unit (Signa HD; GE Medical Systems, Milwaukee,Wis) utilizing a body coil and a 3T MR system (Ingenia, Philips Healthcare, Best, Netherlands) using multiple phased-array surface coils.

The muscle involvement was symmetric in all cases. Muscles of the pelvic girdle and thighs were the most frequently affected, with predominant involvement of the posterior compartment of both thighs and relative sparing of the adductors muscles. All affected muscles showed fatty infiltration on T1

weighted images and no evidence of muscular edema on STIR images. There was no evidence of involvement of upper extremities or thoracic musculature.

Our study indicates that WB MRI may be a useful tool to identify GMPPB mutations in patients with limb-girdle weakness phenotype.

22

Whole-body Muscle MRI in CHRNG-related Multiple Pterigium Syndrome

Daniel Natera-de Benito[1]; Emilio J. Inajeros Clemente[2]; Laura Carrera García[1]; Carlos Ortez[1]; Susana Quijano-Roy[3]; Cristina Jou[1]; Andres Nascimento[1]

[1]Unit of Neuromuscular Disorders, Hospital Sant Joan de Déu, Barcelona (Spain) [2]Department of Radiology, Hospital Sant Joan de Déu, Barcelona (Spain) [3]AP-HP, Hôpitaux Universitaires Paris-Ile-de-France Ouest, Pôle pédiatrique, Centre de Référence Maladies Neuromusculaires (GNMH), Hôpital Raymond Poincaré , Garches; Université Versailles Saint-Quentin UVSQ (France)

The multiple pterygium syndromes (MPS) are a form of arthrogryposis multiplex congenita characterized by the presence of joint contractures (arthrogryposis), pterygia (skin webbing across the joints), and other variable features including short stature, ptosis, low-set ears and cryptorchidism in males. Mutations in CHRNG, which encodes the gamma subunit of the embryonic acetylcholine receptor represent a major cause of MPS.

The purpose of this study is to describe a pattern of muscle involvement in a series of four patients with different clinical phenotypes but all sharing mutations in CHRNG gene.

We followed-up four individuals belonging to three different families (2 females, 2 males) with a mean age of 14.2 year-old (range 7–19) over a mean period of 9 years (range 7–17). All patients carried two CHRNG gene mutations identified by standard procedures. Two patients underwent WB MRI on a 1.5-T MR imaging unit (Signa HD; GE Medical Systems, Milwaukee,Wis) utilizing a body coil and two patients were scanned with a 3T MR system (Ingenia, Philips Healthcare, Best, Netherlands) using multiple phased-array surface coils.

The muscle involvement was symmetric in all cases. Muscles of lower extremities were the most frequently affected. There was a localized pattern of involvement of the fibularis longus and brevis, soleus and tibialis anterior muscles bilaterally, with involvement of the lower erector spinae muscles. T1 weighted images showed fatty replacement of the aforementioned muscles, with no evidence of muscular edema on STIR images. We noted a pseudohypertrophy of latissimus dorsi muscle bilaterally in one patient.

A distinct radiological pattern was identified, with the predominant abnormalities in the muscles of the lower limbs and the lower erector spinae muscles. A major finding of this study is the striking and selective decrease in volume of several muscles, which is not always associated with signal abnormality. This aspect of reduced muscle bulk with a particular combination of muscle volume and signal abnormalities produces a homogeneous and recognizable pattern.

23

Cardiac involvement in Facioscapulohumeral Muscular Dystrophy Type 1 patients with Preserved Ejection Fraction– Assessment by Cardiovascular Magnetic Resonance

Edyta Blaszczyk[1,2] MD, Ulrike Grieben[3] MD, Florian von Knobelsdorff[1] MD, Peter Wellman[4] PhD, Luisa Schmacht[1,2] MD, Stephanie Funk[1,2] MD, Simone Spuler[3] MD, Jeanette Schulz-Menger[1] MD

[1]Working Group on Cardiovascular Magnetic Resonance, Experimental and Clinical Research Center a joint cooperation between the Charité University Medicine Berlin and the Max-Delbrueck Center for Molecular Medicine, and HELIOS Klinikum Berlin Buch, Department of Cardiology and Nephrology, Berlin, Germany [2]DZHK (German Centre for Cardiovascular Research), partner site Berlin [3]Muscle Research Unit, Experimental and Clinical Research Center a joint cooperation between the Charité Medical Faculty and the Max-Delbrueck Center for Molecular Medicine, Berlin, Germany [4]National Heart, Lung and Blood Institute, National Institute of Health

Cardiac involvement in muscle disorders is of growing interest in cardiology. Facioscapulohumeral muscular dystrophy type 1 (FSHD1) is an autosomal dominant disease and is classified on the third place among the most common inherited muscle diseases. The diagnosis of FSHD1 is suspected mainly

in patients with the presence of prominent asymmetric weakness of the face and shoulder muscles. The myocardial involvement in FSHD seems to be underestimated.

The aim of this study is to identify myocardial tissue injury in patients with preserved left ventricular function applying native and contrast-enhanced cardiac magnetic resonance.

Prospectively included patients with a genetically confirmed diagnosis of FSHD1 (n=52, age 48±15 y, Left ventricular ejection fraction, LVEF 63 ± 5%) were compared with 29 healthy age matched controls (p=0.32) using a 1.5 T MR scanner MAGNETOM AvantoFit®, Siemens Healthcare. Myocardial tissue differentiation was performed using fat imaging: multi-echo sequence for fat/water separation, diffuse fibrosis imaging: T1-mapping: MOLLI before and after 0.15mmol/kg bw gadobutrol and for focal fibrosis: late gadolinium enhancement (LGE). ECV was calculated. Analysis was performed using CVI42 (circle cvi).

Focal fibrosis was present in 13 patients (25%,10 men). Fat infiltration was observed in 7 patients (13%,5 men). In healthy volunteers no myocardial injury was detectable. Focal fibrosis positive patients were older (p=0.037), but LVEF was not different (p=0.704). T1 mapping revealed significant differences between all patients and healthy volunteers in global native T1 values basal ($p<0.001$) and medial (p=0.028). In patients, diffuse fibrosis was also detectable in the regions adjacent to focal fibrosis. There were significant sex differences within FSHD1 group with female having more evidence for diffuse injury. Both the global medial T1 values (FSHD1 men:992±26 vs. women: 998±54 ms, p=0.047) and the global medial ECV values (FSHD1 men:26±3 vs. women: 28±3 ms, p=0.028) were increased in females compared to men. No gender differences within healthy were detectable.

In patients with FSHD1 with preserved ejection fraction focal as well as diffuse subclinical myocardial injury is detectable applying CMR. Sex-related differences are assessable within the FSHD1 group applying mapping.

24

Speeding up Clinical Neuromuscular Imaging of the Lower Limbs

Stephen Wastling[1,2], Sachit Shah[1,2], John Thornton[1,2], Tarek Yousry[1,2]

[1]Lysholm Department of Neuroradiology, National Hospital for Neurology and Neurosurgery, Queen Square, London, UK [2]Neuroradiological Academic Unit, Department of Brain Repair and Rehabilitation, Institute of Neurology, UCL, London, UK

In clinical practice, neuromuscular MRI investigations of the limbs are used to guide biopsies, monitor longitudinal changes and to determine the pattern of muscle involvement. Our standard imaging protocol included axial multi-slice spin-echo T1 and T2-STIR weighted imaging performed on 3T Siemens Prisma and Skyra systems. The median duration for an examination of the lower limbs using this protocol was 24 minutes.

To develop a time-focussed neuromuscular MRI protocol for imaging the lower limbs in significantly less time without compromising diagnostic quality.

Imaging parameters in the standard protocol were modified to reduce scan time. By increasing the voxel size from 0.9×0.9×6mm to 1.1×1.1×6mm and reducing the TR from 810 to 792ms, the number of averages from 3 to 1 and the refocussing flip angle from 160 to 120□ the acquisition time of the T1-weighted images was reduced from 6:54 to 2:12. The acquisition time of the T2 STIR images was reduced from 7:04 to 4:12 by reducing number of concatenations from 3 to 2 and increasing the turbo factor from 8 to 11. Two patients were scanned with both the current and time-focussed protocols and the images were reviewed independently by two neuroradiologists (blinded to scan parameters). The overall scan-time savings were then assessed in 40 patient examinations from timing information in the DICOM metadata.

Qualitative review by two neuroradiologists demonstrated no loss of diagnostic quality in the time-focussed images with improved tissue contrast in the STIR images. Parameter changes predicted a scan acquisition time saving of 7 minutes 34 seconds. A median overall scanning time saving of 10 minute 16 second saving per examination was measured. Additional time savings were attributed to reduced planning time because of lower SAR resulting from reduced refocussing flip angles.

A time-focussed protocol was developed that produced diagnostic quality images in a median time of 13 minutes 44 seconds per examination (44% saving). This new protocol will serve as a baseline for comparison as we introduce 3D DIXON imaging.

25

Muscle ultrasound elastography and MRI in preschool children with Duchenne muscular dystrophy

Anna Pichiecchio, MD[1]; Francesco Alessandrino, MD[1]; Chandra Bortolotto, MD[2]; Alessandra Cerica, MD[2]; Raimondo Vitale[2]; Paolo Felisaz[3]; Cristina Rosti, MD[2]; Maria Vittoria Raciti, MD[2]; Marta Rossi, MD[4]; Angela Berardinelli, MD[4]; Giovanni Baranello, MD[5]; Fabrizio Calliada, MD[2] and Stefano Bastianello, MD, PhD[1,5]

[1]Neuroradiology Department, C. Mondino National Neurological Institute, Pavia, Italy [2]Institute of Radiology, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy [3]Ospedale Broni Stradella ASST Pavia [4]Child and Adolescent Unit, IRCCS "C. Mondino" Foundation, Pavia, Italy [5]Developmental Neurology Unit, Istituto Neurologico "Besta" Milan, Milan, Italy [6]Department of Brain and Behavioral Sciences, University of Pavia, Pavia, Italy

Diffuse fibro-fatty changes on T1-weighted (w) muscle MRI scans of Duchenne muscular dystrophy (DMD) patients are widely reported in the literature as well as edematous alterations with short-tau inversion recovery (STIR) sequences. However, without sedation, MRI can be challenging in young children, so other non-invasive imaging methods have been proposed in recent years, including shear wave elastography (SWE). SWE is a ultrasound-based imaging technique that allows quantitative assessment of tissue stiffness and has recently demonstrated higher muscle stiffness in DMD patients than in healthy children (HC).

The aim of this study was to determine muscle tissue elasticity, measured with SWE, in selected lower limb muscles of patients affected by DMD and to correlate the values obtained with those recorded in HC and with muscle MRI data from the same DMD children.

Ten preschool children were enrolled and studied with SWE, five children with a clinical and molecular diagnosis of dystrophinopathy (median age= 48 months) and five age-matched HC. The following inferior limb muscles were studied: gluteus maximus (GM), rectus femoris (RF), vastus medialis (VM), vastus lateralis (VL) and adductor magnus (AM), tibialis anterior (TA) and medial gastrocnemius (MG). DMD patients also underwent muscle MRI of the lower limbs (T1w and STIR sequences). Each muscle was graded according to the degree of fatty replacement and muscle edema using semi-quantitative scales.

In the DMD children, muscle stiffness was significantly increased in the RF (p=0.001), VL (p=0.001), AM (p=0.02) and GM (p=0.02) muscles. On muscle MRI T1-w images showed fatty replacement in 3/5 patients at the level of the GM, while thigh and leg muscles were affected in 2/5; hyperintensity on STIR images was identified in 4/5 patients. No significant correlation was observed between stiffness values and MRI scoring.

Our study demonstrated that lower limb muscles of preschool DMD patients show fatty replacement and patchy edema on muscle MRI and increased stiffness on SWE, findings which are not related to each other. Further studies in larger cohorts are needed to establish whether these changes might be used as follow-up markers.

26

Limb girdle muscular dystrophy due to mutations in POMT2

S.T. Oestergaard[1], K. Johnson[2], T. Stojkovic[3], T. Krag[1], W. De Ridder[4,6], P. De Jonghe[4,6], J. Baets[4,6], K. G. Claeys[7,8], R. Fernandez-Torron[9], L. Phillips[2], A. Töpf[2], J. Colomer[10], S. Nafissi[11], S. Jamal-Omidi[11], C. Bouchet-Seraphin[12], F. Leturcq[13], V. Straub[2], D.G. MacArthur[13,14], M. Lek[14,15], Liwen Xu[14,15], J. Vissing[1]

[1]Copenhagen Neuromuscular Center, Rigshospitalet, University of Copenhagen, Denmark [2]John Walton Muscular Dystrophy Research Centre, Institute of Genetic Medicine, Newcastle University, Newcastle upon Tyne, United Kingdom [3]AP-HP, Institute of Myology, Centre de reference des maladies neuromusculaires Paris Est, G-H Pitié-Salpêtrière, France [4]Neurogenetics Group, Center for Molecular Neurology, VIB, Antwerp, Belgium [5]Laboratory of Neuromuscular Pathology, Institute Born-Bunge, University of Antwerp, Antwerpen, Belgium [6]Neuromuscular Reference Centre, Department of Neurology, Antwerp University Hospital, Antwerpen, Belgium [7]Department of Neurology, Neuromuscular Reference Centre, University Hospitals Leuven, Leuven, Belgium [8]KU Leuven - University of Leuven, Laboratory for Muscle Diseases and Neuropathies, Department of Neurosciences, Experimental Neurology, Leuven, Belgium [9]Neurology Department, Donostia University

Hospital, Neuromuscular Area, Biodonostia Health Research Institute, Donostia-San Sebastian, Spain [10]Unitat de Patología Neuromuscular, Servei de Neurologia, Hospital Sant Joan de Déu, Barcelona, Spain [11]Department of Neurology, Shariati Hospital, Tehran University of Medical Sciences, Tehran, Iran [12]AP-HP, Hôpital Bichat, Département de Biochimie et de Génétique, Paris, France [13]Laboratoire de Génétique et Biologie MoleculairesHopital Cochin, Paris, France [14]Analytic and Translational Genetics Unit, Massachusetts General Hospital, Boston MA 02114, USA [15]Medical and Population Genetics, Broad Institute of MIT and Harvard, Cambridge MA 02142, USA

Mutations in the gene coding for protein O-mannosyl-transferase 2 (POMT2) are known to cause severe congenital muscular dystrophy, but recently, mutations in POMT2 have also been linked to a milder limb-girdle muscular dystrophy (LGMD) phenotype, named LGMD type 2N, but only four cases have been reported so far. Therefore, knowledge about phenotype is scarce.

We aimed to expand the knowledge on degree and pattern of muscle affection, extramuscular symptoms and cognitive involvement in LGMD2N.

We report 12 new cases of LGMD2N, aged 18-63 years. The pattern of muscle involvement was assessed by whole-body muscle MRI in all patients and brain MRI in 10 of the 12 patients. Four cross-sectional slices at the level of calves, thighs, L4 and pelvis were chosen for evaluation of muscle involvement. Replacement of muscle by fat was graded according to the Mercuri scale. Also, we examined muscle strength, clinical features and performed muscle biopsy analysis.

Presenting symptoms were difficulties in walking and running, pain during exercise, delayed motor milestones and learning disabilities at school. All had some degree of cognitive impairment. Brain MRIs were abnormal in three out of ten patients, showing ventricular enlargement in one, periventricular hyperintensities in another, and frontal atrophy of the left hemisphere in a third patient. Muscle MRI imaging revealed a pattern of selective muscle involvement, with most strikingly affected muscles being the hamstring, paraspinal and gluteal muscles. Consistent with the evaluation of muscle strength, the hamstring muscles were more severely affected than the anterior thigh muscle group.

Most affected muscle groups were hip flexors and extensors on strength testing followed by knee flexors and extensors. In the 12 patients of our cohort, 5 alleles were known mutations, while 17 novel mutations accounted for the remaining 19 alleles.

We describe a large cohort of LGMD2N patients, with involvement of hamstrings, but also paraspinal and gluteal muscles on MRI, which correlated well with reduced muscle strength in hip and knee flexors and extensors. Our study showed that LGMD2N is associated with cognitive impairment and significantly expanded the mutational spectrum for LGMD2N, as we describe 17 novel POMT2 mutations in the association with LGMD2.

Abstracts 27-39

New Imaging Techniques

27

Simple semi-automatic method for quantitative analysis of dystrophic muscle biopsies

W.Kloinowski[1], R.Gomolka[1], B.Kuraszkiewicz[1], A.Kaminska[2], P.Stepien[1]

[1]Nalecz Institute of Biocybernetics and Biomedical Engineering PAS, Warsaw, Poland [2]Department of Neurology, Warsaw Medical University, Warsaw, Poland

This is an interdisciplinary work applying methods of biomedical physics and engineering in medical diagnostics.

Recent developments of automated systems for genetic material analysis have caused that nowadays histopathological methods based on images from muscle biopsies are applied not so often as in previous decades since they are more time-consuming . Our aim is to develop simple and quick semiautomatic method of analysi of histopathological images to assist in diagnostics of dystrophies.

Colour microscopic images from biopsies of dystrophic muscles in different stages of Duchenne muscular dystrophy (DMD) and of healthy muscles have been obtained from the archive of the Department of Neurology, Warsaw Medical University. The images were hematoxylin-eosin-stained and clinically evaluated by pathologist specialized in neuromuscular diseases.

Our method is based on colour filtration pixel-by-pixel of the whole microscopic slide and will be called CFPP method. The calculations are easily done in MATLAB. Images are colour-coded in RGB space - each pixel is expressed as a triplet of numbers, (r,g,b), where each number can vary from 0 to 255 (color intensity from darkest to lightest),

We define the connective tissue index (CI)

$$CI = (Lc + Ln) / (Lc + Lm)$$

where Lc, Ln, and Lm denote respectively the numbers of the pixels in the given image belonging to the given class - connective tissue, nuclei, myocytes. For a pixel to be counted into the given class the numbers (r,g,b), must fulfill simple arithmetic inequalities

(Rm <r <RM) AND (Gm <g <GM) AND (Bm <b <BM)

where minimal and maximal values for each colour in the given class are established semi-automatically in MATLAB in such a way that the classes are disconnected in (r,g,b), space. Then using these limit values each image in the given series is analyzed pixel by pixel.

Severity of DMD is assessed by pathologists based on the amount of irregular myocytes, and amount of fatty and connective tissue in the images obtained from biopsies.. Our analyses show that connective tissue index, CI , well correlates with the assessment made by the pathologist.

The proposed method is simple and not time-consuming. It allows to distinguish DMD from normal muscle tissue, as well as potentially gives a possibility to evaluate the severity of DMD.

28

Automated Segmentation of Skeletal Muscle in Dixon MRI of the Thigh based on Texture Features

Rafael Rodrigues and Antonio Pinheiro

Multimedia Signal Processing Group, Instituto de Telecomunicações - Universidade da Beira Interior, Covilhã, Portugal

The segmentation of skeletal muscle is essential for the diagnosis and monitoring of myopathies using anatomical imaging. The manual segmentation of Magnetic Resonance Image (MRI) volumes easily becomes tedious and time-consuming. However, obtaining a fully automated solution is still a challenging task, considering the difficulty in defining proper discriminating visual features in MRI images. Texture analysis has provided interesting results in the characterization of muscle tissue.

In this paper, a fully automated method for the segmentation of skeletal muscle in Dixon sequence MRI images is proposed, based on local texture features classification and using atlas-based information.

The images are preprocessed to obtain a rough binary mask surrounding the muscle area and a binary bone mask, using histogram thresholding combined with morphological filtering and size-based area selection. Image texture is described locally with the Histogram of Oriented Gradients (HOG), the resulting image from the Laplacian of Gaussian (LoG) filter and a local energy descriptor from Haar Wavelet decomposition. Leave-one-out cross-validation is then applied to train AdaBoost with these features and perform pixel-wise classification of 10 Dixon MRI slices. These were selected from volumes of the human thigh, based on largest muscle area criteria. In each cross-validation iteration, ground truth segmentations are used for positive training pixel selection and are also used to build a probabilistic atlas, which serves as a guide for muscle identification on the resulting segmentation. All MRI slices are registered to a common reference before the atlas construction, considering the center of the bone and the convex hull of the ground truth image, or the resulting segmentation in the case of the test MRI.

The obtained segmentations are compared with manual segmentations available for each muscle in the selected Dixon images. Accuracy, precision and recall results will be presented for each muscle, as well as the Dice overlap coefficient.

The described method provides a viable solution for the effective segmentation of skeletal muscle in MRI images, yielding spatially accurate muscle separation.

29

Age-related changes in muscle water diffusivity suggest changes in the sarcolemma during development

Paola Porcari[1], Matt G Hall[2], Chris A Clark[2], Elizabeth Greally[1], Volker Straub[1], Andrew M Blamire[3]

[1]Institute of Genetic Medicine, Newcastle University, UK
[2]UCL GOS Institute of Child Health, London, UK
[3]Institute of Cellular Medicine and Centre for In Vivo Imaging, Newcastle University, UK

During postnatal development, muscle fibres grow enormously and the sarcolemma dynamically and constantly expands. Investigating this process in the time up to maturity may help the understanding of clinical onset of infant myopathies. Diffusion MRI is known to be sensitive to tissue microstructure, and is a promising method for characterising microstructural changes during muscle development.

To investigate the evolution of hindlimb muscle microstructure between young (development) and healthy adult mice using diffusion-weighted imaging protocols and histology.

Male C57BL/10 mice (n=21) from three age groups were investigated using multi-slice-multi-echo T2 images and diffusion-weighted scans (TR/TE=4000/20ms, δ=3ms). Six diffusion times (Δ; range 25-350ms) were explored with diffusion-gradient (Gdiff) applied along or across muscle fibre direction. Apparent-diffusion-coefficients (ADC) were calculated at all diffusion times in the gastrocnemius and tibialis anterior muscles of young (7.5 weeks-old) and adult mice (22/44 weeks-old). Following MRI, mice were injected with Evans Blue Dye (EBD), the percentage of positive uptake determined and excised limbs fixed in formalin. Right hind-limbs underwent imaging using the same protocol as in the in vivo group, with left hind-limbs processed for H&E staining. Feret's diameter of muscle fibres measured using Fiji (Image J).

Hind-limb muscle mean ADC was determined per diffusion time, diffusion-gradient direction and age group. Compared with ADC calculated with Gdiff along fibres, lower values were found with Gdiff across fibres. Muscle water diffusivity was higher in younger than in adult mice where the diffusion behaviour showed no differences between the two investigated ages. In vitro data showed the same diffusivity pattern as in vivo with lower values. For both muscles, the same pattern of muscle fibre size distribution was found histologically across age group. In particular, muscle fibre size increased significantly from young to adult mice whereas the percentage of EBD uptake decreased with age.

This study shows a decrease in water diffusivity between young and mature muscles. Muscle fibre size is larger in adult mice and the extracellular matrix decreases with age. This suggests that diffusivity in young and adult muscles may be driven by different sarcolemma-associated properties between actively growing myofibres and adult myofibres in homeostasis.

30

Microstructural changes in diffusion MRI of muscle tissue in a mouse model of DMD: results from information theoretic entropy

Matt G Hall, Paola Porcari, Elizabeth Greally, Volker Straub, Andrew Blamire, and Chris A Clark

[1]UCL Great Ormond St Institute of Child Health, 30 Guilford St, London WC1E 1EH [2]Newcastle Magnetic Resonance Centre, Newcastle University [3]Institute of Genetic Medicine, Newcastle University, UK

The most common application of MRI to muscle imaging in Duchenne Muscular Dystrophy (DMD) is via fat fraction quantification, providing information about the progress of the replacement of muscle tissue with fat. This technique is powerful, but fat replacement is very much an end-stage of DMD pathology.

Diffusion MRI is known to be sensitive to the tissue environment experienced by spins on length scales far smaller than a typical scan voxel, but challenges remain in how to interpret changes in diffusion decay curves to provide reliable, non-invasive biomarkers. We investigate a model-free approach to diffusion image analysis based on the information theoretic entropy.

Imaging: Using a diffusion-weighted STEAM sequence on a 7T Varian scanner, we acquire images at six diffusion times and four different gradient strengths per diffusion time in three orthogonal directions, one parallel and two perpendicular to muscle fibre direction. TE/TR=4000/20ms, δ=3ms. N=4 wildtype and Mdx mice were imaged.

Histology: The Gatrocnemius muscle was carefully removed from the left fore-limb of each mouse, mounted, frozen, and laminin stained. Feret's diameter was measured using Image-J.

Curve analysis: We calculate the Shannon entropy of the diffusion-weighted data in each voxel, comparing the information content in different regions of the tissue in wild type and Mdx mice.

Entropy maps show contrast across the muscle cross-section and very little noise. Histology reveals a decrease in the mode of the fibre size distribution from wild type to Mdx as well as the Mdx distribution being skewed downwards. The observed entropy of w/t vs Mdx mice shows a significant shift in mean ($1.19\pm0.32 \times 106$ w/t vs. $1.84\pm0.32 \times 106$ Mdx).

Diffusion-weighted measurements acquired over a range of b-values and diffusion times show contrast between wild type and Mdx mice. Entropy provides a potential biomarker which avoids the need for intensive processing or model-fitting. The shift in Entropy of the diffusion-weighted measurements combined with the associated changes in fibre size distribution strongly suggest that diffusion-weighted measurements are sensitive to microstructural changes in muscle. The next step is to apply these approaches in human volunteer and DMD patient data.

31

Texture Analysis of T1 TSE MRI for the Diagnosis and Follow-up of Collagen VI Related Myopathy

Rafael Rodrigues[1], Antonio Pinheiro[1], Susana Quijano-Roy[2,4,5], Robert-Yves Carlier[3,4,5], Benjamin Doré[4], David Gomez-Andrés[6]

[1]Multimedia Signal Processing Group, Instituto de Telecomunicações, Universidade da Beira Interior, Covilhã, PORTUGAL [2]Unité Neuromusculaire pédiatrique, Service de Pédiatrie, Hôpital Universitaire R. Poincaré, Garches, FRANCE [3]Service d'Imagerie Médicale, Pôle Neuro-locomoteur, Hôpital R. Poincaré, Garches, FRANCE [4]Université de Versailles-St Quentin, U1179 UVSQ-INSERM, FRANCE [5]Centre de Référence Maladies Neuromusculaires GNMH, Filière nationale FILNEMUS, FRANCE [6]Child Neurology Unit. Hospital Universitari Vall d'Hebron. Vall d'Hebron Institut de Recerca. Barcelona, SPAIN

Magnetic Resonance Imaging (MRI) is considered as a valuable and noninvasive tool for the assisted diagnosis of Neuromuscular Diseases (NMD). Texture analysis of muscle tissue in MRI has been shown to have a good correlation with the histological changes associated with NMD. Current knowledge suggests the possibility of developing effective models for the description of myopathy based on texture analysis, which could reduce the need for biopsy in both early diagnosis and follow-up of myopathies. However, such models remain an open challenge.

This research focuses on studying variations in texture parameters, considering a variety of myopathy stages, to identify texture features that correlate strongly with MRI outcomes of diagnosed myopathy.

In this work, the authors analyze 23 Whole-body MRI volumes, collected from 21 subjects with a diagnosed (or suspected) myopathy related to Collagen VI (COL6) mutations. The included cases range from mild to severe cases of Ullrich congenital muscular dystrophy and Bethlem myopathy. T1 Turbo Spin Echo (T1 TSE) images of the thigh and upper arm were used. Several regions of interest (ROI) were interactively defined on the thigh muscles and the triceps brachii. These were drawn in similar areas from the muscles in each volume, to enable a meaningful comparison of texture outcomes. From the defined ROI, a set of features based on the Gray Level Correlation Matrix (GLCM), Wavelet analysis, Gabor filters and the Histogram of Oriented Gradients (HOG) were extracted.

Results of the texture analysis were studied and related to the phenotype and severity of COL6 patients. Typical correlation measures (Pearson, Spearman, Kendall) were used to rank computed features based on their discriminative properties.

With this study, the current knowledge on muscle MRI texture analysis is extended. An efficient model for the early diagnosis of Ullrich congenital muscular dystrophy, as well as for assisted follow-up of diagnosed myopathy cases was developed, benefiting from microtexture analysis.

32

Automatic muscle groups segmentation on NMR images based on deep learning techniques

Eduard Snezhko[1], Pierre G. Carlier[2,3], Noura Azzabou[2,3], and Pierre-Yves Baudin[4]

[1]United Institute of Informatics Problems, Minsk, Belarus [2]NMR laboratory, Institute of Myology, Paris, France [3]NMR laboratory, CEA/DRF/IBFJ/MIRCen, Paris, France [4]Consultants for Research in Imaging and Spectroscopy, Tournai, Belgium

Automatic segmentation of muscles in NMR images still remains an open issue, while often being the first step before further quantitative analysis of the segmented muscle groups.

The motivation for this work was to explore the abilities of recent techniques based on Convolutional Neural Networks (CNNs) to segment muscle groups.

Data were acquired on a 3T siemens scanner and using a 3D Gradient-echo sequence with TE= 3.95ms with a pixel resolution of 1x1x5 mm3 and a 448x448x64 matrix size. 184 volumes covering the thighs and 161 for legs were considered. For the training and the test, we performed manual segmentation of the left and right side of following muscle groups: Quariceps, Hamstering, Triceps surae, Extensor and Fibularis. All images were preprocessed to suppress the bias.

For the experiments we utilized a VGG-like CNN (similar to the model by the Visual Geometry Group, Oxford). The network consists of encoder and decoder components. Each component contains four of blocks of layers. Each block of the encoder part contains two or four convolutional + Rectified Linear Unit (ReLU) layers followed by a subsampling layer. The decoder part looks like a symmetrically flipped encoder, where subsampling layers are replaced by upsampling layers. At the end of the decoder part a softmax multi-class classifier is placed.

The networks takes as an input a 2D slice of 448×224 pixels.. For each input pixel it computes the probability of belonging to each muscle group and background.. The pixel is assigned to the muscle group or to the background with highest probability. 80% of volumes were taken for training phase and the remaining were used for testing phase.

Average values of Dice's coefficient of segmentation of the Quadriceps was 0.96 and 0.92 for the Hamstering. Regarding the leg it was 0.9 for the Extensor 0.82 for the Fibularis and 0.90 for the Triceps surae.

Muscle group segmentation using CNNs gave more than acceptable results, even at this very early stage of technical development. Since there is plenty of room for further improvements, the method looks very promising.

33

Spontaneous Mechanical Activities in Healthy Human Leg Musculature Visible in DWI and their Relation to Electrical Activities in EMG

M. Schwartz[1,2], G. Steidle[1], P. Martirosian[1], A. Ramos-Murguialday[3,4], H. Preißl[5,6], A. Stemmer[7], B. Yang[2], F. Schick[1]

[1]Section on Experimental Radiology, University of Tübingen, Tübingen, Germany [2]Institute of Signal Processing and System Theory, University of Stuttgart, Stuttgart, Germany [3]Institute for Medical Psychology and Behavioural Neurobiology, University of Tübingen, Tübingen, Germany [4]Neurotechnology Laboratory, TECNALIA Health Department, San Sebastian, Spain [5]Institute for Diabetes Research and Metabolic Diseases of the Helmholtz Centre Munich at the University of Tuebingen, German Centre for Diabetes Research (DZD), Tübingen, Germany [6]Department of Pharmacy and Biochemistry, Institute of Pharmaceutical Sciences, Interfaculty Centre for Pharmacogenomics and Pharma Research, University of Tübingen, Tübingen, Germany [7]Siemens Healthcare GmbH, Erlangen, Germany

Time series of diffusion-weighted images (DWI) have shown large affected muscle regions impaired by focal spontaneous mechanical activities in musculature (SMAM) (Steidle, 2015).

To get a more detailed insight in muscle physiological processes as expected as underlying functional process of SMAMs in DWI, measurements of DWI and surface electromyography (sEMG) of resting calf musculature were concurrently recorded to reveal spatial and temporal correlation between SMAMs visible in DWI and electrical activities.

Eight healthy volunteers (age: 34±13 years, BMI: 24.8±1.4 kg/m²) were examined after 5 min resting to achieve relaxed musculature. A prototype diffusion-weighted stimulated-echo EPI sequence (Siemens Healthcare GmbH) with matrix size: 64x64, FoV: 192x192 mm², slice-thickness: 6 mm, 6/8 readout, BW: 2004 Hz/px, b-value: 100 s/mm², TE: 31 ms, TR: 500 ms, mixing time TM: 145 ms and diffusion-sensitizing time Δ: 157 ms was applied on a 3 T MR scanner (MAGNETOM Skyra, Siemens Healthcare GmbH). Three bi-polar electrodes for sEMG measurements were placed on the m. gastrocnemius medialis due to expected high activity in this muscle region (Steidle, 2015). sEMG measurements lasted 250 s, respectively 500 repetitions of DWI. sEMG signals were recorded with MR-compatible equipment (BrainAmp ExG MR, Brain Products GmbH) and were MR artifact corrected (Glaser, 2013 and Delorme, 2004). Spontaneous activities were evaluated regarding overall number of occurrences in both modalities and were compared to sEMG measurements outside MR room.

Near-surface SMAMs in a region of 20 mm around electrode location have shown a preceding sEMG event in 84.9±14.3 %. A strong correlation between both modalities was revealed (sEMG events outside MR room and during DWI: 0.9659; sEMG events outside MR room and SMAMs in DWI: 0.9752; sEMG events during DWI and SMAMs in DWI: 0.9750).

Spontaneous activities in a near-surface area are detectable in both modalities enabling studies of their temporal and spatial characteristics. Due to high correlation between both modalities (outside MR room and during DWI), SMAMs should no longer be considered as imaging artifact but rather as new methodology for assessing activity of resting musculature. Changes of activity pattern in subjects with muscular diseases have to be analyzed in future studies.

34

Texture-based segmentation of skeletal muscle in Dixon MRI images

Rafael Rodrigues, Antonio Pinheiro

Instituto de Telecomunicações - UBI, Covilhã, PT

The segmentation of skeletal muscle is essential for the diagnosis and monitoring of myopathies using anatomical imaging. The manual segmentation of MRI volumes easily becomes tedious and extremely time-consuming. However, a fully automated solution is still a very challenging task, considering the difficulty in defining proper discriminating visual features in MRI images. Texture analysis has provided interesting results in the characterization of muscle tissue.

In this paper, we aim at developing an automated method for the segmentation of skeletal muscle in Dixon sequence MRI images. The proposed approach relies on texture analysis through wavelet and gradient based features.

MRI slices were preprocessed to obtain a rough segmentation of the muscle region and the bone, using gray-level gradient and histogram thresholding,

respectively, combined with morphological filtering and size-based area selection. The resulting mask was used both for defining the training pixels and to suppress areas classified as positive outside the muscle region. Image texture was analyzed locally using the histogram of oriented gradients (HOG) descriptor and a reconstructed image using the second level detail components from Haar Wavelet decomposition. Local features such as mean intensity, standard deviation, skewness and kurtosis of both the original gray-level image and the resulting Laplacian of Gaussian (LoG) filtering were also included in the pixel descriptors, which were used to train an AdaBoost classifier. Leave-one-out cross-validation was applied to train AdaBoost and perform pixel-wise classification of a database of 13 Dixon MRI volumes of human thigh scans, considering 40 slices out of each volume. After the slice-by-slice volume segmentation, edge definition was refined using a set of heuristic rules based on information from contiguous slices.

Accuracy, precision and Dice overlap coefficient will be provided in the final paper.

The preliminary segmentation results indicate that texture analysis may provide a viable solution for effective automatic segmentation of skeletal muscle in MRI images.

35

Potential of Stimulated Echo Diffusion Tensor Imaging as disease marker in Duchenne Muscular Dystrophy

Bauke Kogelman[1], Kevin Adamzek[2], Ernst Suidgeest[1], Maaike van Putten[2], Louise van der Weerd[1,2]

[1]C.J. Gorter Center for High Field MRI, Department of Radiology, Leiden University Medical Center, The Netherlands [2]Department of Human Genetics, Leiden University Medical Center, The Netherlands

Duchenne muscular dystrophy (DMD) is a genetic progressive muscle wasting disease, leading to death between the 2nd and 4th decade of life due to cardio-respiratory failure. The DMD mdx mouse model presents with smaller muscle fibers, on average 10-50 microns in diameter, compared to wild type mice. DMD is characterized by early remodeling of muscle microstructure.

Diffusion Tensor Imaging (DTI) can probe muscle microstructure by measuring the amount of water diffusion and may serve as an early disease marker and thus might be more specific than standard T2 measurements. To effectively use diffusion MRI as probe, unconventional long diffusion times (Δ) are required to allow water diffusion reach the regime of restricted diffusion, caused by this muscle microstructure. The aim of this study was to investigate the potential of DTI as an early disease marker.

To study this, 8 week old (n=20) mdx and (n=20) C57BL/10ScSnJ mice were scanned using a 7T Bruker Pharmascan equipped with a 370mT/m gradient system. To explore diffusion hindrance, diffusion tensors were calculated from unweighted and b=500s/mm2 weighted images acquired at Δ=[20-100-200-300-400-500]ms in 8 directions. A T2 map was acquired as well.

Diffusion parameters, mean diffusivity (MD) and lambda 1 to 3, all decreased with increasing diffusion time, while fractional anisotropy (FA) increased. From Δ=20ms to Δ=500ms, MD decreased from 1.232+0.07 to 0.681+0.04 μm2/ms and FA increased from 0.26+0.03 to 0.61+0.04. As expected, T2 values were significantly ($P<0.01$) increased in mdx mice compared to WT mice (34.07+3.09ms vs. 28.03+1.01ms). While T2 values were significantly different, DTI parameters were not.

Lower diffusivities were expected based on smaller muscle fibers in mdx mice, however no significant differences were found in each diffusion time. We hypothesize that the characteristic increased permeability of muscle fibers found in mdx mice, counteracts the effect of restriction by fiber size. Based on the conventional DTI analysis, discrimination between healthy and affected muscle is not possible. We need advanced modelling of diffusion data to further understand the microscopic changes occurring in muscular dystrophies.

36

Quantitative MRI of extra-ocular muscles in the clinical evaluation of systemic diseases

J.W.M. Beenakker[1,2], L. van Vught[1,2], R .de Meel[3], J. Burakiewicz[1], S. Genders[2], I. Notting[2], M. Jager[2], J.J.G.M. Verschuuren[3], H.E. Kan[1]

[1]CJ Gorter center for high field MRI, LUMC, Leiden, Netherlands [2]Department of Ophthalmology, LUMC, Leiden, Netherlands [3]Department of Neurology, LUMC, Leiden, Netherlands

Most imaging techniques within ophthalmology cannot image the extra-ocular tissues. MRI could be extremely useful for this purpose. In Graves orbitopathy, for example, the extra-ocular muscles and orbital fat are inflamed, while in myasthenia gravis, the patients suffer from inability to move the eyes, caused by dysfunction of the extra-ocular muscles. Quantitative MRI data could facilitate a more targeted treatment selection and provide a direct method to determine the treatment response, which varies greatly between patients.

To develop and evaluate the clinical value of the Dixon-technique for systemic ocular conditions.

All scans were performed on a 7Tesla MRI, using a cued-blinking paradigm to minimize eye-motion. The Dixon scan was acquired with a 0.7x0.7x1.0mm3 resolution, TR/TE/FA:2.4ms/10ms/3º.

5 healthy subjects (24-49 years), 3 myasthenia (28-68 years) and 4 Graves orbitopathy (28-64 years) patients were scanned. The muscles, optic nerve and sub-orbital fat were manually segmented on the first coronal slice posterior to the globe. The mean water and fat signal intensity and surface area were determined for all ROIs.

The scans proved to be robust against eye-motion for all participants. The fat fraction of both patient groups was higher than the healthy subjects. The high fat content of the ocular muscles of the myasthenia patients was an unexpected finding and warrants further study. In 5 patients we furthermore performed quantitative T2 mapping, which could add additional clinical information on the amount of muscular inflammation.

Conventional evaluations, such as the clinical activity score, poorly reflect the current condition of the muscles. One patient, for example, was considered to have no functional muscle tissue left, while the MRI showed there was still muscle tissue present, opening new possibilities for treatment.

Quantitative MRI can provide key clinical information on the status of the extra-ocular muscles and orbital fat for patients with systemic diseases, which are not available with the conventional ophthalmic techniques.

37

A Machine Learning Approach for Automated Peripheral Nerve Segmentation

Fabian Balsiger[1], Carolin Steindel[2], Mirjam Arn[2], Benedikt Wagner[2], Marwan El-Koussy[2], Mauricio Reyes[1], Waldo Valenzuela[1,2], and Olivier Scheidegger[2,3]

[1]Institute for Surgical Technology and Biomechanics, University of Bern, Switzerland [2]Support Center for Advanced Neuroimaging (SCAN), Institute for Diagnostic and Interventional Neuroradiology, Inselspital, Bern University Hospital, University of Bern, Switzerland [3]Department of Neurology, Inselspital, Bern University Hospital, University of Bern, Switzerland

Quantitative NMR techniques of peripheral nerves have received low attention in the neuromuscular scientific community lately. Approximately 2.4 % of the population suffer from peripheral neuropathy, whose diagnosis relies on neurological examination and electrodiagnostic studies (EDS). Nerve segments close to the trunk remain difficult to assess using these techniques. Nowadays, magnetic resonance neurography (MRN) is increasingly used as a complementary diagnostic tool, which enables physicians to image deeply situated nerves and its surrounding tissues.

We hypothesize that quantitative measures such as volume and shape, calculated from segmented peripheral nerves, can assist in the diagnosis and monitoring of peripheral neuropathies. Therefore, we aim to develop a peripheral nerve segmentation method from MRN images.

We propose a fully-automatic multi-sequence segmentation pipeline consisting of five main steps: (i) pre-processing using bias field correction, histogram matching, and smoothing, (ii) registration of the turbo inversion recovery magnitude to T2-weighted MRN sequence, (iii) extraction of various features including intensity and anatomical location-based descriptors, (iv) voxel-wise tissue classification into peripheral nerve and background using a machine learning model based on decision forests, and (v) post-processing to obtain a final segmentation of the peripheral nerves. We evaluated the performance on a cohort of six volunteers with MRN images of the upper leg using a leave-one-out cross-validation strategy and the Dice coefficient as evaluation metric.

The automated segmentation yields a Dice coefficient of 0.788 ± 0.054. Qualitatively, the overall

shape of the obtained 3-D nerve reconstructions is consistent with the manually segmented ground truth. In regards to the machine learning model, an analysis of the feature importance showed that the strongest descriptors of nerve agree with the visual cues used by physicians when locating and delineating peripheral nerves.

These preliminary results suggest that the proposed method is able to fully-automatically segment peripheral nerves in the upper leg with a good accuracy. Future work includes the improvement of our method by using deep learning techniques and the segmentation of peripheral nerves in other regions than the upper leg. Moreover, we will evaluate the performance of our algorithm on cohorts of patients who have been diagnosed with peripheral neuropathy by EDS.

38

Real-time MRI (RT-MRI) for evaluation of dysphagia in inclusion body myositis

Per-Ole Carstens*[1], Arno Olthoff*[2], Shuo Zhang[3], Eva von Fintel[4], Joachim Lotz[4], Jens Frahm**[3], Jens Schmidt**[1]

/ Authors contributed equally to this work.*

[1]Department of Neurology, University Medical Centre Göttingen, Göttingen, Germany; per-ole.carstens@med.uni-goettingen.de; j.schmidt@med.uni-goettingen.de
[2]Department of Otorhinolaryngology, Phoniatrics and Pedaudiology, University Medical Centre Göttingen, Göttingen, Germany; olthoff@med.uni-goettingen.de
[3]Biomedizinische NMR Forschungs GmbH at the Max Planck Institute for Biophysical Chemistry, Göttingen, Germany; szhang1@gwdg.de, jfrahm@gwdg.de [4]Institute of Radiology, University Medical Centre Göttingen, Göttingen, Germany; joachim.lotz@med.uni-goettingen.de, eva.vonfintel @med.uni-goettingen.de

Inclusion body myositis (IBM) is a rare acquired myopathy that is typically observed in patients above 50 years. The disease is characterized by a progressive muscle weakness and dysphagia.

The aim of this study was to compare the assessment of dyphagia by real-time magnetic resonance imaging (RT-MRI) to standard assessment by flexible endoscopic evaluation of swallowing (FEES) and videofluoroscopy (VF) in a cohort of patients with IBM and to assess safety and feasibility of this technique in patients with dysphagia.

Using RT-MRI, FEES and VF, swallowing was studied in 20 unselected and consecutive patients with IBM as the index disease. Symptoms of dysphagia and IBM were explored by standardized tools including Swallowing-Related Quality of Life Questionnaire (SWAL-QoL), IBM Functional Rating Scale, and Medical Research Council Scale.

Swallowing in a supine position during RT-MRI was well-tolerated by all patients and no complications like aspiration or dyspnea occurred. RT-MRI was non-inferior in the assessment of retentions compared to standard examination by VF and FEES. Only RT-MRI allowed precise time measurements of oral and pharyngeal transportation times and identification of the respective tissue morphology. The pharyngeal transit time was significantly prolonged compared to published reference values and significantly correlated with morphologic abnormalities. Dysphagia was noted in 80% of the patients.

The technique of RT-MRI for the assessment of dysphagia is safe and equally capable as VF to identify the cause of dysphagia in IBM. Advantages of RT-MRI include a more exact measurement of transportation times during swallowing, a superior visualization of soft tissue, and lack of X-ray exposure. RT-MRI may become a routine diagnostic tool for detailed assessment of the esophagus, facilitating longitudinal evaluations in daily practice and clinical trials in a wide range of myopathic and other neurological disorders.

39

Brain imaging indicates genotype-phenotype association in Duchenne muscular dystrophy

Nathalie Doorenweerd[1,2], Chiara Marini Bettolo[2], Kieren G. Hollingsworth[3], Jos G.M. Hendriksen[4,5,7], Erik H. Niks[6,7], Volker Straub[2], Hermien E. Kan[2]

[1]John Walton Muscular Dystrophy Research Centre, Newcastle University, Newcastle Upon Tyne, United Kingdom [2]C.J. Gorter Center for High Field MRI, Department of Radiology, Leiden University Medical Center, Leiden, The Netherlands [3]Newcastle Magnetic Resonance Centre, Institute of Cellular Medicine, Newcastle University, Newcastle upon Tyne, United Kingdom [4]Kempenhaeghe Epilepsy Center, Department of Neurological Learning Disabilities, Heeze, The Netherlands [5]Department of Neurology, Maastricht University Medical Center, Maastricht, The Netherlands [6]Department of Neurology, Leiden University Medical Center, Leiden, The Netherlands [7]Dutch Duchenne Centre, The Netherlands

Duchenne muscular dystrophy (DMD) is associated with specific learning and behavioural disabilities. Using quantitative MRI, we previously reported reduced grey matter volume, altered white matter microstructure and reduced cerebral blood flow in DMD patients compared to healthy age-matched controls. Patients missing multiple dystrophin isoforms, Dp140 in addition to the full length Dp427, had the lowest volume, lowest perfusion and largest changes in white matter.

To determine whether patients with mutations in the DMD gene that lead to missing all dystrophin isoforms, including Dp71, were even more affected.

3D T1-weighted images were obtained on a 3T Philips system using an 8-channel head coil at two different sites. Scans from 36 DMD patients (8-21 years old, steroid naïve n=4), 34 healthy controls (8-29 years old) and six limb-girdle muscular dystrophy 2I (LGMD2I) patients (12-28 years old) were processed using FSL software to calculate intracranial, total brain (TBV), grey matter (GMV), white matter (WMV) and CSF volumes. A short neuropsychological examination was performed that focussed on specific cognitive tasks known to be at risk in DMD. DMD patients were subdivided into three groups based on the mutation location within the DMD gene (Dp427 n=16, Dp427-Dp140 n=15, Dp427-Dp71 n=5).

Smaller TBV (Dp427 -2%, Dp427-Dp140 -7%, Dp427-Dp71 -12%) and GMV (Dp427 -5%, Dp427-Dp140 -7%, Dp427-Dp71 -14%) was found compared to healthy controls. DMD patients in group Dp427 and LGMD2I patients did not differ significantly from the healthy controls, whereas DMD Dp427-Dp140 and Dp427-Dp71 did ($p<0.01$). All patients in group Dp427-Dp71 had low scores on their neuropsychological tests and 4/5 patients were diagnosed with general developmental delay.

A cumulative loss of dystrophin isoforms coincided with lower TBV and GMV, as well as increased behavioural difficulties and worse cognitive performance. These results are in line with clinical findings, where patients lacking more isoforms are at greater risk for developing cognitive and behavioural problems.

Abstracts 40-80

Quantitative Muscle Imaging

40

3D Architecture of Human Lower Leg Muscles Assessed with Ultra-High-Field Diffusion Tensor Imaging and Tractography: Sensitivity to Sex Difference and Intramuscular Variability

Alexandre Fouré[1], Augustin Ogier[1], Arnaud Le Troter[1], Christophe Vilmen[1], Thorsten Feiweier[3], Maxime Guye[1,2], Julien Gondin[4], Pierre Besson[1], David Bendahan[1]

[1]Aix Marseille Univ, CNRS, CRMBM, UMR 7339 [2]APHM, Hôpital de La Timone, CEMEREM [3]Institut NeuroMyoGène, Université Claude Bernard Lyon 1, INSERM U1217, CNRS 5310 [4]Siemens Healthcare

Neuromuscular disorders induce skeletal muscle function impairments related to changes in both muscle volume and structural arrangement of fascicles/fibers within the muscle leading to the reduction of skeletal muscle force production. However, the estimation of both muscle volume and architecture in a single experimental session remains challenging.

To demonstrate the sensitivity of the lower leg muscles 3D structural organization measurements using diffusion tensor imaging (DTI) assessed with ultra-high-field (7T) MRI and tractography of skeletal muscle fibers.

Intramuscular variability and sex difference were characterized at 7T on lower leg muscles of young healthy men (n = 10) and women (n = 10). Isometric voluntary plantar flexion force was assessed to establish potential correlation with muscle geometric characteristics. Student t-test, statistical parametric mapping and Spearman coefficient (ρ) were used for statistical data analysis.

Significant sex differences were detected in muscle volume (+ 21.7 % in men for the entire lower leg, P = 0.008) while architecture parameters were almost identical across sex. Additional differences were found independently of sex in architecture along several muscles of the lower leg. A significant correlation was established between the physiological cross-sectional area of the main plantar flexor muscles and isometric voluntary force capacities (ρ = 0.56, P = 0.011).

The high-resolution DTI assessed with 7T-MRI allows a reproducible assessment of muscle microarchitecture and structural organization of superficial and deep muscles giving indirect information on muscle function. Theses parameters can help the diagnosis of muscular alterations and the determination of potential clinical interventions effects in patients with muscular disorders and in injured athletes.

41

Reproducibility of DTI parameters over a 2 week period in the hamstrings of healthy athletes

Jithsa Monte[1], Melissa Hooijmans[1], Jos Oudeman[1], Martijn Froeling[2], Johannes Tol[4], Gustav J. Strijkers[3], Mario Maas[1], Aart J. Nederveen[1]

[1]Academic Medical Center – University of Amsterdam, Department of Radiology & Nuclear Medicine [2]Utrecht Medical Center – University of Utrecht, Department of Radiology [3]Academic Medical Center – University of Amsterdam, Department of Biomedical Engineering and Physics [4]Academic Medical Center – University of Amsterdam, Department of orthopedics

Muscle injuries are the most common injuries in team sports, with the highest incidence for hamstring injuries. Conventionally and widely used T2-weighted imaging techniques are insufficient for assessing tissue repair and predicting return to play. Diffusion Tensor imaging (DTI) can probe muscle fiber micro-architecture and has shown to be sensitive to muscle changes that remained undetected on conventional T2-weighted MRI. However, little is known about the normal variability in DTI parameters in uninjured hamstring muscles.

Determine the reproducibility in DTI parameters over the course of 2 weeks in uninjured hamstring muscles.

DTI datasets (FOV 252x480mm; TR/TE 5914/55ms; 12 gradient directions; b-value 400 s/

mm2; voxel size 3x3x5mm; no gap; 40 slices) were acquired in the upper legs of five healthy athletes (age 26.8±7.5 yrs range: 22-40) with a 2 week interval, using a 3T MR system (Ingenia; Philips Healthcare, Best, the Netherlands). DTI datasets were processed using DTITools for Wolfram Mathematica. Mean diffusivity (MD), fractional anisotropy (FA), λ1, λ2 and λ3 were calculated for each muscle at both time points (Biceps femoris long head (BFLH), Semimembranosus (SB), Semitendinosus (ST)). The mean value was calculated over the 20 middle slices. Muscles with an SNR<20 at b0 were excluded from the analysis. The statistical analysis comprised Bland-Altman plots and coefficient of variation (CV).

25 out of 30 muscles were included in the analysis. Low CV-values and good agreement in the Bland-Altman analysis were found between the measurements. The CV values were:

all muscles: 4.9% for λ1 , 3.5% for λ2, 3.6% for λ3, 5.2% for MD and 3.8% for FA;

BFLH: 5.2% for λ1 , 3.2% for λ2, 3.4% for λ3, 3.9% for MD and 5.8% for FA;

ST: 5.1% for λ1 , 4.4% for λ2, 3.9% for λ3, 4.5% for MD and 3.4% for FA;

SB: 4.2% for λ1 , 3.2% for λ2, 4.0% for λ3, 1.9% for MD and 5.4% for FA.

High reproducibility for all DTI parameters was found, indicating that the DTI method is reproducible and might be sensitive to detect minimal changes in acute muscle injuries.

42

Examining the relationship between Dixon quantitative MRI and Physiotherapy functional outcome measures in Dysferlinopathy

Roberto Fernandez-Torron[1,2], Meredith K. James[1], Helen Sutherland[1], Fiona E. Smith[3], Noura Azzabou[4], Ian Wilson[3], Harmen Reyngoudt[4], Anna Mayhew[1], Andrew Blamire[3], Pierre G Carlier[4], Heather Hilsden[1], Laura Rufibach[5], Marni Jacobs[6,7],The Jain COS Consortium, Kate Bushby[1], Volker Straub[1]

[1]John Walton Muscular Dystrophy Research Centre, Newcastle upon Tyne, UK [2]Neuromuscular Area. Biodonostia Health Research Institute. Neurology Service. Donostia University Hospital. Donostia-San Sebastian. Spain [3]Magnetic Resonance Centre, Institute for Cellular Medicine, Newcastle University [4]AIM & CEA NMR Laboratory, Institute of Myology, Pitié-Salpêtrière University Hospital, 47-83, Paris, France [5]The Jain Foundation, Seattle, Washington, USA [6]Center for Translational Science, Division of Biostatistics and Study Methodology, Children's National Health System, Washington, DC, USA [7]Pediatrics, Epidemiology and Biostatistics, George Washington University, Washington, DC, USA

Dysferlinopathy presents with a spectrum of muscle weakness including distal and proximal phenotypes and variable rate of progression. This variability presents challenges for developing clinical trial outcome measures. The Jain COS Consortium aims to overcome these challenges by collecting observational data from 203 genetically confirmed patients (aged 12-88) over 3 years.

The aim of this piece of work is to investigate the relationship between Physiotherapy measures and Dixon quantitative MRI data over the first year of the 3-year international Clinical Outcome Study of Dysferlinopathy.

Patients underwent physiotherapy, medical and MRI assessments. Physiotherapy assessments included muscle strength (manual muscle testing; hand held dynamometry) and functional ability evaluations (North Star Assessment for Dysferlinopathy and Performance of Upper limb), as well as timed tests (rise from floor, 10 metre walk / run, four stair climb and descend; Timed Up and Go, Six Minute Walk Distance) and respiratory function testing. Quantitative MRI on the lower limbs was completed at baseline and one year using the Dixon technique for fat/ water separation.

Fat fraction values were obtained for all muscles in the lower legs using a region of interest based approach and the change between baseline and year one assessed using Wilcoxon's Signed Rank Tests. Significance was set at $p < 0.05$. Data evaluation at this point included MRI from 100 patients. The Physiotherapy data collected at baseline and one year visits were analysed to calculate the median change for the cohort. We examined the relationship between the fat fraction and Physiotherapy data. The data were examined to identify groups demonstrating differing rates of progression. We report which muscles are most affected and examine the relationship of change between fat fraction and functional outcome data.

This work shows that progression in Dysferlinopathy can be demonstrated using both quantitative fat fraction and functional outcomes over one year.

43

Comparison of whole leg muscle versus individual muscle fat fraction values in a multi-centre natural history study of Dysferlinopathy at year 2 follow up

F.E. Smith[1], D.R. Al-Asadi[1], I. Wilson[1], R. Fernandez Torron[3], N. Azzabou[2], H. Reyngoudt[2], H Sutherland[3]; H. Hilsden[3], A. Blamire[1], P. Carlier[2], The Jain COS Consortium; K. Bushby[3], V. Straub[3]

[1]NMR Centre, Institute of Cellular Medicine, Newcastle University, Newcastle upon Tyne, UK [2]Institute of Myology, Paris ,FR [3]The John Walton Muscular Dystrophy Research Centre, Institute of Genetic Medicine, Newcastle University, Newcastle upon Tyne, UK

Dysferlinopathy is a rare neuromuscular disorder caused by mutations in the dysferlin gene leading to progressive skeletal muscle wasting. The International Clinical Outcome Study of Dysferlinopathy is studying the natural history of this disease over a three year period.

To monitor disease progression, we considered fat fraction (FF) as a biomarker. Here we report whole leg FF results versus individual muscle values from baseline to year 2 of the 4 year study.

203 subjects (11 to 86 years old) were enrolled across 14 centers (Europe, USA, Australia, Japan). This work reports findings in a subset of 70 subjects selected from the entire cohort. Scans were acquired from different systems at 1.5T and 3T. Lower limbs were imaged using gradient-echo Three-point Dixon (2D or 3D) with optimized TEs (echo time) and TR: (repetition time), and with TR between 10ms and 100 ms and flip angle between 3° and 10°. Thigh and leg muscles were manually delineated to obtain the mean FF value per muscle over each year. A mean FF was also determined for the whole thigh and lower-leg muscles. The change of FF in each muscle and in the whole leg muscles were assessed using non-parametric Wilcoxon tests, a Spearman′s correlation was performed to compare individual thigh and leg muscle FFs with whole muscle values.

Results At baseline, the FF was higher in all thigh muscles compared to the lower leg muscles by at least 7%. FF significantly increased each year by an average of 5% for thigh muscles and was most significant in the anterior compartment (p0.8) with whole leg values.

All individual thigh FFs increased significantly over each year. Whole lower-leg and thigh FF values were highly correlated with individual muscle values. FF, as a biomarker is useful for monitoring disease progression over time as well as therapeutic efficacy in future clinical trials, with whole leg values appearing as sensitive as individual muscles to detect disease progression.

44

Statistical modeling of 5-year longitudinal data from a large cohort of Duchenne muscular dystrophy (DMD) subjects: an interim look at temporal characteristics of disease progression from the ImagingDMD study

William D. Rooney, Yosef Berlow, Sean C. Forbes, Rebecca J. Willcocks, William T. Triplett, Dah-Jyuu Wang, Barry J. Byrne, Richard Finkel, Barry S. Russman, Elliot O'Brien, Brendan Moloney, Erika L. Finanger, Michael J. Daniels, H. Lee Sweeney, Glenn A. Walter, and Krista H. Vandenborne.

[1]Advanced Imaging Research Center, Oregon Health & Science University, Portland, Oregon, United States [2]Department of Physical Therapy, University of Florida, Gainesville, Florida, United States [3]Physiology and Functional Genomics, University of Florida, Gainesville, Florida, United States [4]Department of Radiology, Children's Hospital of Philadelphia, Philadelphia, Pennsylvania, United States[5], Department of Pediatrics, University of Florida, Gainesville, Florida, United States [6]Department of Neurology, Nemours Children's Hospital, Orlando, Florida, United States [7]Shriners Hospital, Portland, Oregon, United States [8]Department of Statistics, University of Florida, Gainesville, Florida, United States [9]Department of Pharmacology and Therapeutics, University of Florida, Gainesville, Florida United States.

Magnetic resonance imaging (MRI) and spectroscopy (MRS) biomarkers are sensitive to Duchenne muscular dystrophy (DMD) disease progression, are non-invasive, and can readily be deployed at multiple sites to facilitate rapid testing of new therapies. The resulting data sets are extensive and complex, so techniques to extract and summarize key features are desirable.

The aim of this study was to investigate modeling approaches of MRI/MRS data sets to generate

efficient summaries of temporal characteristics of DMD disease progression in individual subjects.

Data were acquired from 211 male subjects aged 5-18y at study entry comprised of 157 DMD subjects and 54 healthy controls. Lower and upper leg data were collected using 3T MRI instruments at three institutions. The DMD subjects were followed at yearly intervals for up to five years. 1H MRS data were acquired using a single voxel spectroscopy sequence to estimate fat fraction (FF) in vastus lateralis and soleus muscles. MRI quantitative T2 (qT2) values were determined from a multi-slice multiple spin echo sequence, and analyzed for eight muscle groups. Longitudinal changes in FF and qT2 were modeled at the group and individual levels using a non-linear mixed effects (NLME) approach with a fitting kernel comprised of a 4-parameter continuous distribution function (CDF).

DMD average FF and qT2 values increased annually for all muscle groups. Group average behavior with age and was well-represented using a CDF. NLME CDF modeling was found to be stable and robust as applied in this setting. Significant heterogeneity in disease progression was observed between muscle groups, and also between individuals. The group average maximum disease progression rate constant varied substantially (up to 3x) between muscles; greatest in biceps femoris long head and lowest in tibialis posterior. Within a given muscle, progression rate constants varied markedly (up to 4x) between individuals. MRI qT2 values were strongly associated with MRS FF. NLME results from FF and qT2 of the same muscle were nearly identical with no significant differences in parameters returned.

NLME CDF modeling provides a robust and reliable approach to quantify temporal aspects of DMD muscle involvement.

45

Diagnostic value of muscle MRI in a cohort of 150 patients from the John Walton Muscular Dystrophy Research Centre

Katja Storch[1]; Volker Straub[1]; Shona Cameron[1]; Chiara Marini Bettolo[1]

[1]John Walton Muscular Dystrophy Research Centre, Institute of Genetic Medicine, Newcastle University, Newcastle upon Tyne UK

The use of muscle magnetic resonance imaging (MRI) in patients with genetic neuromuscular conditions has increased over the last decade. This is due to an improved understanding of selective patterns of muscle involvement in these rare conditions. Muscle MRI can be a useful diagnostic tool to direct genetic testing and therefore lead to more efficient diagnosing.

The aim of this review is to evaluate the diagnostic value of muscle MRI in genetic muscle disease.

We selected a cohort of 150 patients from the John Walton Muscular Dystrophy Research Centre t who had a muscle MRI to investigate if the a particular pattern of muscle involvement contributed to the decision on genetic testing. Furthermore, we wanted to find out which specific genes were targeted based on the differential diagnosis suggested by MRI.

In 30 % (45) of patients the established genetic diagnosis was supported by the selective pattern of muscle involvement seen on MRI scans, which were performed after the genetic diagnosis had been established. In all except one case, the MRI showed a typical pattern of the underlying disease.

In 40.7% (61) of patients a muscle MRI was performed in undiagnosed patients to help direct genetic testing. In 39.3% (24) of these the suspected diagnosis based on muscle MRI was confirmed genetically. The diseases correctly identified by muscle MRI analysis were: Bethlem myopathy (5), FSHD (1), RYR1-associated myopathy (5), Pompe disease (1), inclusion body myopathy (2), Myofibrillar myopathy (1), limb-girdle muscular dystrophy (LGMD) type 2A (3), 2B (4), 2L (1), and STIM-1 related myopathy (1).

In 42.6 % (26) of patients imaged, the MRI did not lead to a diagnosis. 31.1% (19) of patients remained undiagnosed. 11.5 % (7) have since been diagnosed with subsequent gene testing. In 4 of these cases different form of LGMD was diagnosed. In 16.4 % (10) testing was not performed or results were not available yet.

Our results suggest that muscle MRI is a valuable diagnostic tool in the workup of patients with rare genetic neuromuscular diseases. In nearly 40 % of cases a genetic testing was correctly guided by muscle MRI.

46

Improved multi-component muscle T2 mapping using maximum likelihood estimation

N Zafeiropoulos[1,2], RL Janiczek[3], CDJ Sinclair[1,2], L Zampedri[2], U Klickovic[2], Fratta P[2], J Morrow[2], M Hanna[2], L Greensmith[2], TA Yousry[1,2], E De Vita[1], Fratta P[2], JS Thornton[1,2]

[1]Neuroradiological Academic Unit, UCL Institute of Neurology, London, UK [2]MRC Centre for Neuromuscular Diseases, UCL Institute of Neurology, London [3]GlaxoSmithKline, London, UK

CPMG T2 relaxometry, sensitive to muscle-water and fat-faction (ff) changes in neuromuscular diseases, is commonly implemented by fitting exponential functions to the multi-echo signal using least-squares (LSQ) minimization. However processes besides simple exponential transverse-magnetization decay contribute to the signal e.g. stimulated and alternate echo effects. Also, the assumptions of normally distributed and homoscedastic noise implicit in LSQ minimization often break down in reality.

To address these limitations by combining the recently-introduced extended phase graph (EPG) approach accurately modelling the signal, with maximum likelihood estimation (MLE) considering the Rician MRI noise distribution.

Simulations generated 500 EPG signal replicates for various realistic multi-component ground-truth sequence and tissue parameter combinations. Muscle-T2 (T2m) and ff estimates were compared for the EPG-MLE and EXP-LSQ fitting methods. Preliminary validation was obtained by comparison with 25-pixel regions-of-interest in vivo results.

Estimator bias was reduced for EPG-MLE vs. EXP-LSQ, e.g. for simulated healthy-muscle conditions (ground-truth T2m 39ms & ff 8%) with excitation flip angle (phi) 60 degrees and SNR 55.5, estimated T2m was 39.1 ± 2.5 vs. 38.4 ± 2.3ms (mean ± SD), and ff 7.8 ± 2.5 vs. 10.9 ± 1.6% respectively. In comparable in vivo data, estimated T2m was 38.9 ± 1.0 vs. 36.3 ± 0.7ms and ff 9.2 ± 4.0 vs. 11.8 ± 3.1% respectively.

For lower flip-angle conditions, e.g. phi = 35 degrees with resulting SNR = 24, EPG-MLE yielded a greater reduction in estimator bias, e.g. for the same ground-truth T2m and ff, estimated T2m was 39.4 ± 6.9 vs. 54.1 ± 8.1ms, and ff 7.3 ± 4.9 vs. 26.2 ± 6.5% respectively in simulation. In similar in vivo conditions estimated T2m was 37.4 ± 4.7 vs. 57.4 ± 8.1ms, and ff 4.4 ± 2.0 vs. 17.3 ± 2.7% respectively.

Under conditions of phi >60 degrees, EPG-MLE and EXP-LSQ yielded comparable values for T2m. EXP-LSQ tended to overestimate ff.

EPG-MLE improves CPMG T2m and ff estimation. The trade-off between accuracy and computational cost is the subject of current work. The next stage will be application in patient studies assessing T2m and ff as neuromuscular disease trial outcome measures.

47

Age and strain related T2 differences in Sgcg(-/-) mice

Ravneet Vohra[1], Stephen Chrzanowski[2], Abhinandan Batra[3], Elizabeth Barton[4], Glenn Walter[2]

[1]Department of Radiology, University of Washington, Seattle, WA, USA [2]Department of Physiology and Functional Genomics, University of Florida, Gainesville, FL, USA [3]Department of Physical Therapy, University of Florida, Gainesville, FL, USA [4]Department of Applied Physiology and Kinesiology, University of Florida, Gainesville, FL, USA

Mutation in the genes encoding sarcoglycan proteins lead to muscular dystrophy in humans and mouse models. The dystrophic phenotype can be highly variable even in the presence of identical gene mutations. This variability, in part, has been attributed to presence of genetic modifiers. Magnetic resonance imaging (MRI) has emerged as a sensitive non-invasive biomarker to monitor the disease progression in both skeletal and cardiac muscles in muscular dystrophies.

To monitor the disease progression in skeletal and cardiac muscles of gamma-sarcoglycan (Sgcg-/-) mice on C57 BL6 (g-BL6) and DBA (g-DBA) background.

Hind limb muscles of three and nine months old, g-BL6 and g-DBA; n = 5 mice were imaged using 4.7T horizontal bore magnet (Agilent VMJ version 3.1). 1H2O spectroscopic relaxometry was implemented using a single voxel with in posterior muscle compartment using stimulated echo acquisition mode (STEAM) under the following parameters: voxel size = 1.5x3.0x1.5 mm3; TR = 9,000 msec; 29 unequally spaced TE's from 5 to 200 msec (5, 6, 7, 8, 9, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 80, 90, 100, 110, 120, 130, 140, 150, 160, 170, and

200); mixing time (TM) = 20 ms and number of phased cycled averages = 4. Water amplitude as a function of TE were determined using complex PCA as previously described 50. The decay in water signal amplitude was fit to a single exponential decay.

T2 using spectroscopy revealed age related changes in skeletal muscles of g-DBA mice. T2 decreased significantly in both dystrophic strains with age (g-bl6; young vs old; mean +/- SD; 29.8 +/- 2.8 vs 25.7 +/- 1.7 ms, g-dba; 24.4 +/- 1.0 vs 22.5 +/- 0.5 ms). Old g-dba mice displayed reduced T2 compared to age matched g-bl6 mice (22.5 +/- 0.5 vs 25.7 +/- 1.7 ms).

Dystrophic mice on dba background demonstrate increased skeletal muscle pathology compared to BL/6 background. MRI was able to differentiate between dystrophic mouse models of separate genetic background.

48

Respiratory muscle composition and breathing dynamics assessed by MRI in Duchenne muscular dystrophy

Alison Barnard, PT, DPT[1]; Donovan Lott, PT, PhD[1]; Abhinandan Batra, MA, PT[1]; Rebecca Willcocks, PhD[1]; Sean Forbes, PhD[1]; William Triplett, BS[1]; Samuel Riehl, BS[1]; Barbara Smith, PT, PhD[1]; Krista Vandenborne, PT, PhD[1]; Glenn Walter, PhD[2]

[1]Department of Physical Therapy, University of Florida
[2]Department of Physiology and Functional Genomics, University of Florida

Duchenne muscular dystrophy (DMD) is a progressive muscle wasting disease, and respiratory impairment contributes to significant morbidity and mortality. Traditional pulmonary function tests are valuable in monitoring respiratory impairment, but they do not allow for differentiation of the distinct factors contributing to respiratory decline.

The aim of this study is to use MRI to investigate accessory respiratory muscle composition and breathing dynamics in DMD.

Nine controls (6-18yrs) and 20 individuals with DMD (8-18yrs) underwent an MRI exam on a 3T Phillips Achieva scanner using a 32 channel cardiac coil. The exam included 3 point Dixon imaging of the accessory respiratory muscles (TR=10ms, TE= 5.4, 6.4, 7.4ms, flip angle = 3 and 10 degrees) and CINE imaging of the chest in the sagittal plane during quiet and maximal breaths. Fatty infiltration of the expiratory muscles (fat fraction = FF) was quantified from reconstructed fat and water images. Diaphragm and chest wall movement were quantified from CINE imaging. Participants also performed forced vital capacity, maximal inspiratory pressure (MIP), and maximal expiratory pressure (MEP) tests.

Participants with DMD had significantly elevated expiratory respiratory muscle FF compared to controls with FF=0.30±0.17, p=0.002 in the rectus abdominis; FF=0.37±0.25, p=0.001 in the external oblique, and FF=0.47±0.28, p<0.001 in the internal oblique. MEP was negatively correlated to the average FF of the expiratory muscles (r=-0.67, p=0.007). Qualitatively, the intercostal muscles demonstrated minimal fatty infiltration and relative sparing in DMD, except in one participant with advanced disease. Using sagittal CINE imaging, we found that in controls, chest expansion was responsible for 58% of lung expansion during maximal inspiration, while in DMD, chest expansion was only responsible for 38% of lung expansion (p=0.012).

The high degree of fatty infiltration in the expiratory muscles is consistent with clinical findings of early expiratory muscle weakness and impaired cough. The decreased ability to expand the chest wall during maximal inspiration may indicate that chest wall compliance limitations exist prior to overt intercostal or diaphragm weakness, although this hypothesis needs further testing. These findings help contribute to a better understanding of the natural history progression of respiratory function in DMD.

49

Quantitative Imaging of the gluteus maximus in spinal cord injured and healthy subjects

Jules L. Nelissen[1,2], Jitsha R. Monte[3], Maurits Sloots[4], Dorien Verschuren[1,2], Larry de Graaf[1], Klaas Nicolay[1], Ralph Sinkus[5], Jurgen H. Runge[3,5], Chistof Smit[4,6], Thomas W. J. Janssen[4,6], Cees W. J. Oomens[7], Aart J. Nederveen[3], and Gustav J. Strijkers[2]

[1]Biomedical NMR, Eindhoven University of Technology, Eindhoven, Netherlands [2]Preclinical and Translational MRI, Academic Medical Center, Amsterdam, Netherlands [3]Radiology, Academic Medical Center, Amsterdam, Netherlands [4]Amsterdam Rehabilitation Center | Reade, Amsterdam, The Netherlands [5]Division of Imaging Sciences & Biomedical Engineering, King's College London, London, United Kingdom [6]Faculty of Behavioural and Movement Sciences, Vrije Universiteit Amsterdam, Amsterdam, The Netherlands [7]Biomechanics of Soft Tissues, Eindhoven University of Technology, Eindhoven, Netherlands

Impairment of the gluteus maximus muscle function in spinal cord injured (SCI) subjects is accompanied with compositional changes, including atrophy, fat deposition, fibrosis, and edema. In combination with impaired or absent sensibility to perceive high soft tissue loads, SCI subjects are extremely susceptible (prevalence of 85% throughout life) for developing pressure ulcers (PrU). Skeletal muscle tissue health is thought to be essential as preventive measure for PrU development.

Investigate health status of the gluteus muscle of healthy and SCI subjects with quantitative Magnetic Resonance Imaging.

2 SCI subjects (SCI-001: ♂, C5, 1997, PrU (18x) and SCI-002: ♂, T9, 2007, PrU (2x)) and 15 healthy volunteers (11♂, 10♀) were included in this study. Muscle stiffness Gd was measured with Magnetic Resonance Elastography (MRE). T2-mapping was performed to measure muscle water T2. DIXON water and fat images were acquired for fat fraction FFdix determination. All measurements were performed with a 3T MRI scanner (Philips Healthcare, Best, The Netherlands). MRE, T2, and DIXON data was processed to obtain quantitative maps. Region-Of-Interest based analysis was performed on a central imaging volume of 8 mm thickness in which the gluteus maximus was selected. Mean T2, Gd and FFdix of the ROI were determined.

All healthy subjects showed comparable T2, FFdix and Gd. Average muscle T2 was 28±4 ms, FFdix was 5.1±1.8%, and Gd was 0.75±0.06kPa over all 15 healthy subjects. A clear difference between healthy and SCI subjects was found. In addition, both SCI subject differed substantially from each other. SCI-001 showed severe muscle atrophy and fat infiltration, whereas in SCI-002 the muscle volume was comparable to healthy volunteers and little fat infiltration was visible. For both SCI subjects the determined mean values were: SCI-001: T2=36±17ms, FFdix=54.9±9.7%, Gd=0.53±0.12kPa. SCI-002: T2=33±3ms, FFdix=13.5±7.4%, Gd=0.53±0.15kPa.

Quantitative Magnetic Resonance Imaging can provide additional information for characterization of the muscle health status and could provide insights in the susceptibility of individual SCI subjects for the development of pressure ulcers.

Acknowledgement

This research was supported by the Dutch Technology Foundation STW (project 12398) and COST BM1304 MYOMRI action. The authors thank Sandra van den Berg – Faay for technical assistance.

50

1H NMRI and 31P NMRS in skeletal muscle of dysferlinopathy patients: 1 year follow-up results in multi-center trial

Noura Azzabou[1,2], Harmen Reyngoudt[1,2], Fiona E. Smith[3,4], Andrew Blamire[3,4], Pierre G. Carlier[1,2], The JAIN Foundation Consortium

[1]NMR laboratory, Institute of Myology, Paris, France [2]NMR laboratory, CEA/DRF/IBFJ/MIRCen, Paris, France [3]Newcastle Magnetic Resonance Centre, Newcastle University, Newcastle upon Tyne, UK [4]The John Walton Muscular Dystrophy Research Centre, Newcastle University, Newcastle upon Tyne, UK

Dysferlinopathy is a rare neuromuscular disorder caused by mutations in the dysferlin gene leading to progressive skeletal muscle wasting. NMR imaging (NMRI) and phosphorus spectroscopy (31P NMRS) can be used as noninvasive quantitative tools in longitudinal studies of dysferlinopathy.

Our objective was to monitor the disease activity and progression over a one year period by assessing, respectively, water T2 and fat fraction (FF). Phosphorus metabolism was also evaluated.

203 subjects were enrolled across 14 centers. Here, we report the results of a patients subset (n=106 between 11 and 86 years old) that underwent successful NMR exams within a year interval. Lower limbs were scanned using two main sequences: (1) Multi-Slice Multi-Echo with echo-train length from 14 to 17 echoes (depending on site), echo times ranging from 9 to 200 ms and a repetition time between 2500 and 4000 ms. (2) Gradient-echo 3-points Dixon with TEs optimized for field, and with TR between 10 and 100 ms and flip angle between 3° and 10°. In 3 centers (26 patients with good SNR data), 31P-NMRS was performed with an FID and/or a 2D-CSI sequence at the level of the tibialis anterior muscle. Changes in NMR indices were compared in each muscle using a non-parametric Wilcoxon test.

Regarding water T2, no significant changes were observed over the course of one year. The FF was higher in thigh muscles (46.8%) than in the leg (41.8%). FF significantly increased by 3 % in average for both the leg and the thigh muscles. The FF increase was significantly higher in subjects with increased water T2 values (>39ms) at both visits when compared to subjects with an increased water

T2 at only one visit and those with no increased water T2. The 31P-spectroscopy biomarkers did not show significant changes over one year; however, we observed a splitting of the Pi resonance in several patients.

Results in this large cohort of dysferlin-deficient patients showed essentially a stable disease activity and progression of skeletal muscle lesions. Benchmarking natural history data is a mandatory step for the future use of NMR biomarkers in order to assess future treatment efficacy.

51

Optical and Magnetic Resonance Imaging of Dystrophic Muscle

Stephen Mark Chrzanowski[1], Ravneet Vohra[1], Brittany Lee-McMullen[2], Abhinandan Batra[3], Ray Spradlin[4], Jazmine Morales[4], Sean Forbes[3], Krista Vandenborne[3], Elisabeth R. Barton[4], Glenn Walter[1]

[1]Department of Physiology and Functional Genomics, University of Florida, Gainesville, FL [2]Department of Biochemistry, University of Florida, Gainesville, FL [3]Department of Physical Therapy, University of Florida, Gainesville, FL [4]Department of Applied Physiology and Kinesiology, University of Florida, Gainesville, FL

Quantification of muscle pathology is a critical outcome measure to assess the efficacy of clinical trials studying muscular dystrophies. Unfortunately, invasive means of assessment are utilized to assess quantitative muscle pathology. Magnetic resonance imaging (MRI) provides valuable information, but is timely and expensive, suggesting the need for an inexpensive, time-efficient, non-invasive means of quantitatively assess muscle pathology. Indocyanine Green (ICG) enhanced near infrared (NIR) optical imaging offers an objective, minimally invasive, and longitudinal modality that to quantify muscle pathology.

The aims of this study were to determine if ICG enhanced NIR optical imaging could be used to a) differentiate dystrophic (mdx and gsg-/-) murine muscle from healthy muscle, b) assess the exacerbation of damaged muscle in dystrophic muscle, and c) measure.

6-10 week old control, mdx, and gsg -/- mice were cross-sectional compared by NIR optical imaging, MRI, histology, and spectrophotometry. Additionally, before and after downhill treadmill running, data were collected from a cohort of mdx mice. Finally, a subset of gsg -/- mice received intramuscular injections of recombinant human SGCG, with data collected before and 6 weeks after the injections. 2D fluorescence images were captured and MRI-T2 relaxation was calculated for comparison. Following imaging, standard histological techniques and spectrophotometry were performed.

Both dystrophic mouse models demonstrated elevated radiant efficiency and MRI-T2 values compared to control counterparts. Eccentric induced muscle damage, via downhill treadmill running, resulted in elevated radiant efficiency and MRI-T2 values. Finally, administration of recombinant human SGCG (desAAV8hSGCG) resulted in decreased ICG fluorescence and MRI-T2 values compared to pre-intervention and non-treated counterparts.

NIR optical imaging is comparable to MRI and can be used to detect muscle damage in dystrophic muscle, monitor worsening of muscle pathology in muscular dystrophy, and assess regression of following therapeutic intervention in muscular dystrophies in a cost effective, safe, and non-invasive manner.

52

Quantitative muscle ultrasound analysis in FSHD patients

Di Pasquale A[1], Fragiotta G[2],Vanoli F[1], L. Licchelli[1], De Santis T[1], Fionda L[1], Garibaldi M[1], Bucci E[1], Vizzaccaro E[1], MorinoS[1], Grossi A[1], Tupler R[2], Antonini G[1]

[1]Department of Neurological Sciences, Mental Health and Sensory Organs (NESMOS) "Sapienza" University of Rome [2]Department of Medico-Surgical Sciences and Biotechnologies "Sapienza" University of Rome Polo Pontino [3]Department of Life Sciences, University of Modena and Reggio Emilia

Muscular imaging represents an important diagnostic tool for the identification and quantification of muscular alterations in patients affected by muscular disorders. Although MRI represents the gold standard to investigate the muscular structure, ultrasound (US) still remains a valid diagnostic tool in the diagnosis of myopathies. Muscle thinning and increase in echo intensity caused by fibro-adipose substitution, are both pathological hallmarks of myopathy. However, considering the variability of such parameters, US diagnostic reliability is still debated.

The aim of our work is to evaluate the applicability of quantitative muscular ultrasonography (QMUS) in the study of myopathies, particularly in patients affected by facioscapulohumeral dystrophy (FSHD).

Until now we evaluated 4 FSHD1 patients and 4 healthy controls matched by sex, age and BMI. Clinical severity was evaluated by FSHD score. With a linear transducer (7-14 MHz) we bilaterally scanned the pectoralis major, deltoid, rectus femoris, tibialis anterior and semimembranosus muscles. In each image QMUS was performed calculating the mean muscle echo intensity by computer assisted gray-scale analysis, keeping gain, depth, number of focuses and transducer pressure constant.The same muscles were evaluated with MRI and muscle's alterations were categorized as normal, altered in T1 or in STIR sequences. QMUS evaluator was blinded to MRI data.

80 muscles were evaluated (40 from patients and 40 from controls). Muscles of FSHD patients displayed a significant echogenicity increase compared to controls ($p \leq 0.001$). Patients with lower FSHD score presented a significantly lower muscles echogenicity, compared to patients with higher scores ($p \leq 0.001$). A good level of correspondence was found between QMUS and MRI evaluation, in particular, muscles with T1 alterations were more hyperechogenic, while muscles altered in STIR sequences were more hypoechogenic compared to normal muscles.

Muscular US allows a qualitative and quantitative evaluation of muscle echogenicity. In FSHD patients, QMUS displayed a tight correlation with the anatomic muscular alterations. In the limited clinical records analysed, it seems that the US data also correlate with the clinical and MRI data. Although a confirmation on larger sample is needed, our research suggests a possible future application of QMUS in the diagnosis of muscular disorders.

53

Interactive segmentation of leg muscles in NMR images

Pierre-Yves Baudin[1], Morin Beyeler[2], Pierre G Carlier[4], Olivier Scheidegger[2]

[1]Consultants for Research in Imaging and Spectroscopy, Tournai, Belgium [2]Support Center for Advanced Neuroimaging, Institute for Diagnostic and Interventional Neuroradiology, Inselspital, Bern University Hospital, University of Bern, Switzerland [3]NMR Laboratory, Institute of Myology, Paris, France [4]NMR laboratory, CEA/DRF/IBFJ/MIRCen, Paris, France

In today's clinical practice and therapeutic trials on neuromuscular diseases, quantitative NMRI is progressively taking a major role for monitoring diseases evolution and treatments efficacy.

However, when processing the acquired data, one typical obstacle lies in the determination of the regions to assess. In studies focusing on the brain or the heart, specialized software tools can automatically segment anatomical images into meaningful regi

A software dedicated to interactive segmentation of muscle NMRI has been introduced recently. It was made available to a team of neurologists who tested it on the lower leg muscles of healthy volunteers and Duchenne patients, the latter presenting severe fatty infiltrations. The resulting ROI were compared to manually drawn ones both in terms of accuracy and processing time. Twelve lower leg muscles were systematically evaluated. All images were acquired either on a 3 tesla Siemens scanner and the acquisition sequence was a standard 2pt Dixon sequence.

The mean segmentation time with the proposed method was 2min per muscle on the volunteer cohort, while it was 35min with the manual approach, yielding a speed increase by a factor of 18. On the Duchenne patients, the acceleration factor was also considerable. The fatty infiltrations did not hinder the algorithm performance.

These results showed that satisfying segmentations could be obtained in a fraction of the time normally taken with the standard approach, while demonstrating the robustness of the software to muscle fatty infiltration. As confirmed by visual inspection, all delineated regions were deemed usable for further analysis.

54

Characterization of pH dysregulation in skeletal muscle of Duchenne muscular dystrophy patients using 31P and 1H nuclear magnetic resonance spectroscopy

Harmen Reyngoudt, Suna Turk, Pierre G. Carlier

[1]NMR laboratory, Institute of Myology, Paris, France
[2]CEA/DRF/IBFJ/MIRCen, Paris, France

It has long been established that skeletal muscle pH of Duchenne muscular dystrophy (DMD) patients is systematically measured as abnormally alkaline by 31P nuclear magnetic resonance spectroscopy (NMRS). A splitting of the inorganic phosphate (Pi) peak, with a second Pi resonance (Pi,b) found downfield from Pi,a, was present both in skeletal muscle of Golden Retriever muscular dystrophy dogs and in DMD patients. This alkaline phosphate pool may originate from leaky dystrophic myocytes or an increased interstitial space.

1H NMRS, exploiting the pH-sensitive proton resonances of carnosine, an intracellular dipeptide, was used to distinguish between these two hypotheses.

NMR data were obtained in 20 DMD patients and in 7 control subjects on a 3-T Siemens Prisma clinical NMR system. Both 31P and 1H NMRS data were acquired at the level of the gastrocnemius medialis muscle. A multi-slice multi-echo imaging (MSME) acquisition was performed for the determination of water T2 and fat fraction, using a tri-exponential fit, in the same region of interest.

31P NMRS-derived pH values were systematically increased in DMD patients, whereas this was not the case for 1H NMRS-based pH. Interestingly, the carnosine-based intracellular pH was never found alkaline in the absence of a concurrent Pi-based pH elevation. Also, abnormal intracellular pH, based on carnosine, was hardly ever associated with normal water T2 values.

The combined 1H and 31P approach used here identified that the two proposed mechanisms can exist. In muscles where 1H and 31P pH estimates are in agreement, it reflects an intracellular origin, more specifically an ionic dysregulation in dystrophic myocytes. In the other DMD patients, intracellular pH was normal but an alkaline Pi pool was still present, suggesting an extracellular origin, likely revealing an expanded interstitial volume fraction, often associated to fibrotic changes. This data supports that 1H NMRS could possibly serve as a biomarker to assess normalization of intramyocytic pH and sarcolemmal permeability following therapy that induces dystrophin expression in DMD patients.

55

Fast T1-mapping for monitoring chronic fatty degenerations in the skeletal muscles of dystrophic patients

Benjamin Marty[1,2], Bertrand Coppa[1,2], Pierre-Yves Baudin[3], Pierre G. Carlier[1,2]

[1]Institute of Myology, NMR Laboratory, Paris, France
[2]CEA, DRF, I2BM, MIRCen, NMR Laboratory, Paris, France [3]Consultants for Research in Imaging and Spectroscopy, Tournai, Belgium

Recent years have seen tremendous progress towards the development of quantitative NMR-based outcome measures to monitor NMD. Methods based on the chemical shift difference between water and fat protons are currently the reference approaches to quantitatively assess fat infiltration in muscle tissues. T1 values are also strongly affected when healthy muscle is chronically replaced by fat. With the development of fast imaging, quantitative T1 imaging is now possible within short acquisition times.

In this study, we present a fast T1 mapping sequence dedicated to skeletal muscles and compared the sensitivity of this approach with a more standard Dixon method to monitor fatty infiltrations within the course of a NMD.

Imaging was performed on the thighs of 10 healthy volunteers and 30 patients suffering from Becker muscular dystrophy (BMD). The T1 mapping sequence consisted in the acquisition of a 1000 radial spokes FLASH echo train following a single magnetization inversion. The following parameters were used: TE/TR = 2.75/5.08ms, resolution = 1.8x1.8mm2, 5 slices, Tacq = 50s. Temporal image series were reconstructed with view-sharing and a compressed sensing algorithm from which T1 maps were derived using Bloch simulations and a dictionary fitting approach. Fat fractions were also quantified using a standard 3D GRE acquisition and the 3-pt Dixon approach.

T1 values were significantly lower in the skeletal muscles of BMD patients compared to healthy volunteers (885±344ms VS 1199±45ms, $p<0.05$), while

fat fraction were significantly higher in patients (37.0±32.7% VS 5.1±3.0 %, $p < 0.05$). In the patient group, T1 values negatively correlated with fat fractions ($R = -0.98$, $p < 0.05$). A few FF maps reconstructed from the standard Dixon presented the classical water/fat swapping artifact while T1 maps were usable for all subjects.

The sensitivity of the T1 mapping sequence was equivalent to the standard water/fat separation method for discriminating between healthy and dystrophic conditions. The short acquisition time of 10 seconds per slice allows dynamic acquisitions or quantitative imaging on pediatric populations while being highly sensitive to FF variations. In conclusion, quantitative T1 mapping is a good candidate for fast, sensitive and quantitative monitoring degenerative changes in NMDs.

56

Quantitative muscle ultrasonography in facioscapulohumeral dystrophy

Rianne JM Goselink[1], Merel Jansen[2], Nicol C Voermans[1], Corrie E Erasmus[1], Baziel GM van Engelen[1], Tim Schreuder[1], Nens van Alfen[1]

[1]Department of Neurology, Donders Center for Neuroscience, Radboud university medical center, Nijmegen, The Netherlands [2]Department of Rehabilitation, Donders Center for Neuroscience, Radboud university medical center, Nijmegen, The Netherlands

Objective, relevant and patient friendly biomarkers are highly needed in FSHD, especially in upcoming clinical trials tailored to reduce muscle dystrophy. Muscle ultrasonography assesses the structural muscle changes without being influenced by fatigue or cooperation and can thus quantify the pathology underlying deterioration without invasive techniques.

The aim of this study is to assess muscle ultrasonography as a biomarker in patients with FSHD. Here, we present data from the baseline regarding the validity of muscle ultrasonography in FSHD.

We performed a prospective study on 25 patients with genetically confirmed and clinically affected FSHD. Five muscle (trapezius, biceps brachii, rectus abdominis, rectus femoris and tibialis anterior muscle) were bilaterally screened and compared with clinical outcome measures (FSHD evaluation score, clinical severity score, and manual muscle testing).

Participants with varying age (10-67 years, mean 40 years) and varying severity (2-15, mean 5) had mildly to severely abnormal quantitative ultrasound measures which showed reasonable to good correlation to a range of clinical parameters.

Muscle ultrasound appears to be a valid surrogate outcome measure, reflecting severity of muscle weakness in patients with FSHD. Muscle ultrasound images can be easily and reliably obtained after minimal training and can be executed with a standard ultrasound apparatus with a linear probe with 3-12 mHz.

57

Muscle MRI for Disease Progression in MND

Klickovic U, Zampedri L, Sinclair C, Shah S, Clarke J, Howard R, Malaspina A, Orrell R, Sharma N, Sidle K, Hanna M, Golay X, Yousry T, Morrow J, Greensmith L, Thornton J, Fratta P.

UCL Institute of Neurology, and MRC Centre for Neuromuscular Diseases

Novel therapies for motor neuron disorders (MND) require reliable and reproducible outcome measures to monitor disease progression. Muscle magnetic resonance imaging (MRI) is an excellent candidate due to its reproducibility and observer independence. Limited MRI data in motor-neuropathies have been published to date, mainly targeting the corticospinal tract and motor cortex, as well as muscle volumetrics in peripheral nerve disorders. No longitudinal muscle MRI studies in patients with motor-neuropathies have been published to date.

In this ongoing clinical study we aim to evaluate the use of skeletal muscle MRI as a biomarker over 12 month period in patients suffering from MND, but having variable rates of disease progression: amyotrophic lateral sclerosis (ALS) and spinal and bulba.

3 study groups [SBMA patients (n=20), ALS patients (n=20) and healthy controls (n=18)] have undergone 3-Tesla MRI scanning of lower limbs, upper limbs, and bulbar region at baseline. 6 and 12 months follow-up examinations are ongoing. The protocol includes semi-quantitative descriptive rating of T1-w images, and quantitative 3-point Dixon and T2-relaxometry MRI to assess muscle fatty infiltration, oedema and atrophy. MRI findings will be correlated over time with functional and clinical assessments in all 3 study groups.

Widespread intramuscular fat accumulation, predominantly in the SBMA patient group can be observed using both, semi-quantitative and quantitative muscle MRI approaches at baseline, with correlations of muscle fat fraction with corresponding clinical and functional assessments. We expect similar associations to be observed in the longitudinal analysis.

At present, clinical trials in MND rely on binary outcome measures such as number of patients surviving at a specified time point. Therefore, establishing biomarkers providing surrogate outcome readouts is an important step for the development of new treatments. Our study will establish the potential of muscle MRI as an outcome marker in motor-neuropathies.

58

Muscle MRI in a large cohort of patients with oculopharyngeal muscular dystrophy

Kroon, H.M.J.M.[1]; Horlings, C.G.C.[2]; de Swart, B.J.M.[1]; Kalf, J.G.[1], van Engelen, B.G.M.[2]

[1]Department of Rehabilitation, Radboud University Medical Centre, Nijmegen, Netherlands [2]Department of Neurology, Radboud University Medical Centre, Nijmegen, Netherlands

Oculopharyngeal muscular dystrophy is a rare late-onset muscular dystrophy often presenting with ptosis and swallowing difficulties, but also limb muscle weakness. This study presents muscle imaging data of a large cohort of OPMD patients.

To quantitatively measure muscle fatty infiltration using muscle MRI in a large cohort of patients with oculopharyngeal muscular dystrophy.

Fifty OPMD patients and family members participated. All were genetically tested for OPMD. Mean age 60 years of age (range 44-79), 22 men, 26 women. We performed muscle MRI of head-neck region and lower extremities. We quantitatively assessed muscle fatty infiltration using DIXON of the lower leg muscles. All patients were clinically assessed using muscle force measurements, motor function measure and chewing and swallowing tests.

Muscle MRI showed muscle fatty infiltration (defined as >30%) in at least one muscle in 40 patients, and 21 patients showed fatty infiltration of at least 30% in one or more muscles. Most affected muscles were soleus, hamstrings and adductor muscles. Fat fractions ranged from <10% (normal) to almost 90% in some muscles. M. rectus femoris did not show fat fractions above 20%, except in 2 severely affected patients. Patients reporting leg muscle involvement, also showed greater fatty infiltration of leg muscles. Some patients showed muscle fatty infiltration, but no clinical symptoms of muscle weakness.

This is the largest study on quantitative muscle imaging in OPMD patients thus far, showing a variable range of fatty infiltration, also in patient with no clinical symptoms.

59

Development of a training and evaluation programme for manual muscle segmentation

Morrow JM[1], Shah S[2], Evans MRB[1], Wastling S[2], Emira AK[2], Klickovic U[1], Machado P[1], Reilly MM[1], Hanna MG[1], Thornton JT[2], Yousry TA[2]

[1]MRC Centre for Neuromuscular Diseases, UCL Institute of Neurology, London, UK [2]Neuroradiological Academic Unit, UCL Institute of Neurology, London, UK

MRI quantification of intramuscular fat fraction has been shown to be a sensitive measure of disease progression in neuromuscular diseases but is usually reliant on manual segmentation of regions of interest (ROI) for analysis. In large clinical trials longitudinal reliability is of importance and multiple observers may be necessary.

We aimed to develop a programme such that new observers were adequately trained and could demonstrate competence prior to commencing muscle segmentation of new datasets.

Previously acquired three-point Dixon acquisitions and derived fat fraction maps at thigh and calf level from patients with neuromuscular disease and healthy controls were utilised for the programme. The training consisted of an instruction manual for ROI segmentation, a training set of eight scans with gold standard ROI defined for reference, and access to an experienced observer for input as required. The assessment involved defining ROI on a single limb at thigh (10 ROI) and calf (6 ROI) level on a pre-specified slice for repeat scans at a two week interval in four subjects (two patients and two controls). Three experienced observers undertook the assessment such that acceptable test-retest and inter-observer reliability of both muscle cross sectional area

and fat fraction could be determined. New observers were assessed against these standards.

For all three experienced observers, test-retest intra-class correlation (ICC) was greater than 0.98 for cross-sectional area and greater than 0.995 for muscle fat fraction at both thigh and calf level. Inter-observer ICC was initially more variable, identifying differing interpretation of the muscle segmentation instructions between these observers. This was used to refine the instruction manual and update the regions of interest. Inter-observer ICC was then greater than 0.93 for cross-sectional area and greater than 0.98 for fat fraction for pairwise comparisons of all three observers at both thigh and calf level. To date one new observer has been successfully trained and assessed to these standards.

We have developed a programme for training and evaluating new observers in manual muscle segmentation of thigh and calf muscles and defined the minimal level of competence for this programme in terms of both inter-scan and inter-observer ICC.

60

Water T2 mapping of the fatty infiltrated thigh musculature using a T2-prepared 3D TSE sequence combined with SPAIR

Sarah Schlaeger[1,2], Dominik Weidlich[1], Elisabeth Klupp[2], Marcus Deschauer[3], Benedikt Schoser[4], Sarah Bublitz[3], Federica Montagnese[4], Ernst J. Rummeny[1], Claus Zimmer[2], Jan S. Kirschke[2], Dimitrios C. Karampinos[1]

[1]Department of Diagnostic and Interventional Radiology, Technical University of Munich, Munich, Germany [2]Department of Diagnostic and Interventional Neuroradiology, Technical University of Munich, Munich, Germany [3]Department of Neurology, Technical University of Munich, Munich, Germany [4]Friedrich-Baur-Institut, Ludwig-Maximilians-University, Munich, Germany

Muscle water T2 mapping can provide useful information about monitoring disease activity in patients with neuromuscular diseases. 2D multi-echo spin-echo (MESE) sequences have been typically used to measure muscle T2 with known limitations including their sensitivity to transmit B1 inhomogeneities and the effect of fat on T2 quantification. The extended phase graph formulation has been proposed in order to model the effect of transmit B1 inhomogeneities and fat on T2. Alternatively, a 3D TSE sequence employing an adiabatic modified BIR-4 T2 preparation of variable duration (T2Prep 3D TSE) and spectral adiabatic inversion recovery (SPAIR) for main fat peaks suppression has been recently shown to minimize the T2 sensitivity to transmit B1 inhomogeneities at 3T in healthy thigh muscles.

To compare the muscle T2 using 2D MESE and T2Prep 3D TSE against the muscle water T2 from MRS as a gold standard in the thigh muscles of patients with neuromuscular diseases.

2D MESE and T2Prep 3D TSE were performed in 9 patients with varied neuromuscular diseases at 3T, using SPAIR in both sequences. Spectra were obtained in healthy (n=9), fatty (n=6) and edematous (n=7) muscles using a STEAM MRS with variable TEs. T2 water and fat fraction values were extracted from the spectra. The MRS water T2 was compared to the imaging muscle T2.

There was a poor agreement of the MESE T2 with the MRS water T2 ($R2=0.31$, $p=0.16$). A good agreement was reported between the T2Prep 3D TSE T2 and the MRS water T2 ($R2=0.98$, $p < 0.01$) with a slope/intercept of 1.13/-1.08ms. The mean relative error of MESE T2 compared to MRS water T2 was 86.8%, whereas the mean relative error of T2Prep 3D TSE T2 compared to MRS water T2 was 9.3%.

The T2Prep 3D TSE sequence results in T2 values in good agreement with water T2 values. The 2D MESE sequence overestimates the T2 values especially in regions with high fat fractions, probably due to the influence of the unsuppressed olefinic fat peak. This effect of the olefinic fat peak may be reduced in T2Prep 3D TSE due to its faster T2 decay.

61

Quantitative muscle analysis of the qualitative MRI images of patients with Duchenne muscular dystrophy

Akihiko Ishiyama[1,4], Ayaka Hirasawa[1], Takahiro Nakayama[2], Satoshi Kuru[3], Hirofumi Komaki[1], Ichizo Nishino[4], Masayuki Sasaki[1]

[1]Department of Child Neurology, National Center Hospital, National Center of Neurology and Psychiatry, Kodaira, Tokyo, Japan [2]Department of Neurology, Division of Neuromuscular Diseases, Yokohama Rosai Hospital [3]Department of Neurology, National Hospital Organization, Suzuka Hospital, Suzuka, Mie, Japan [4]Department of Neuromuscular Research, National Institute of Neuroscience, National Center Hospital, National Center of Neurology and Psychiatry, Kodaira, Tokyo, Japan

The signal of pixels on muscle MRI was considered to be proportional to the ratio of muscle to fat, so we determine the muscle MRI value and the fat MRI value as constants, and create the formula of the impairment ratio. The impairment ratio of the muscle was calculated from the average pixel values of the each muscle. The ratio increases with increasing fat content.

We use the automatic calculate muscle impairment ratio system to analyze muscle MRI of Duchenne muscular dystrophy (DMD) patients.

Muscle MRI of the pelvis, thigh and calf muscles taken from forty-four boys with DMD ranging from one to ten years old (mean: 4.75 years) was used. We measured the pixel value of each muscle DICOM image of T1-weighed MRI using Image/J, calculated muscle impairment ratio and assessed the progression and variation of fatty infiltration of each muscle.

In pelvis and thigh muscles, the gluteus maximus and adductor magnus had the highest impairment ratios in every age, and semimembranosus, semitendinosus, and biceps femoris followed these ratios. In calf muscles, gastrocnemius had the highest. We created a scatter diagram of relationship between age and impairment ratio and the regression line (regression equation: y= ax+b). The gluteus maximus and biceps femoris had the highest regression coefficient (a=0.2885, 0.1833), these were followed by rectus femoral and adductor magnus (a=0.1672, 0.1571). On the other hand sartorius and gracilis had the lowest. (a=0.0067, 0.0092). These dates indicate that the fatty infiltration of the gluteus maximus and adductor magnus muscles occurred earlier and the sartorius and gracilis were relatively preserved.

Our study coincides with many of previous, and demonstrated that auto calculation of muscle impairment ratio is useful for evaluating the degree of distribution of muscle fatty infiltration quantitatively. There is no special skill to use this automatic calculation system, so it would be a powerful tool for monitoring disease progression and efficacy of treatment strategies.

62

MR imaging in Spinal Muscular Atrophy as a biomarker for disease progression. Protocol design for an observational cohort study with 1 year follow-up.

Louise AM Otto MD[1], Martijn Froeling PhD[2], Lara V Schlaffke PhD[2,3,4], Wieke Haakma MSc[2], Marloes Stam MD[1], Camiel A Wijngaarde MD[1], Bart Bartels MSc[5], Leonard H van den Berg MD; PhD[1], Jeroen Hendrikse MD; PhD[2], W Ludo van der Pol MD; PhD[1]

[1]Brain Centre Rudolf Magnus, Department of Neurology, University Medical Centre Utrecht, Utrecht, The Netherlands [2]Department of Radiology, University Medical Centre Utrecht, Utrecht, The Netherlands [3]Department of Neurology, BG-University Hospital Bergmannsheil, Ruhr-University Bochum, Bochum, Germany [4]C.J. Gorter Centre for High Field MRI, Department of Radiology, Leiden University Medical Centre, Leiden, The Netherlands [5]Department of Child Development and Exercise Centre, University Medical Centre Utrecht, Utrecht, The Netherlands

Recently developed treatment strategies for hereditary proximal spinal muscular atrophy (SMA) have further increased the urgency for sensitive clinical outcome measures. Biomarkers that are able to capture treatment efficacy or disease progression before it is reflected by a deterioration of motor function are warranted.

To investigate the value of DIXON, T2 mapping and diffusion tensor imaging (DTI) as quantitative magnetic resonance imaging (MRI) biomarkers for assessing SMA severity and monitoring disease progression.

Study population: 30 patients with genetically confirmed SMA, types 2 or 3; 30 age-matched healthy controls without motor neuron diseases or myopathy, aged >12 years. SMA patients will be recruited from the Dutch SMA database of the SMA Centre of Expertise at the University Medical Centre of Utrecht, The Netherlands. Exclusion criteria consist of any type of (non-)invasive ventilation, severe swallowing disorders, >15% discrepancy in FVC due to postural changes from sitting to supine, or any contra-indication for 3-T MRI.

Study design: A 15 cm image stack (FOV 48x24 cm2) of both upper legs will be acquired on a 3-T MRI scanner (Philips Ingenia) of all patients and healthy controls. The MRI protocol will take 10

minutes and consists of 1) Dixon Imaging (6 point multi acquisition FFE Dixon; ΔTE 0.76 ms; vox: 1.5x1.5x6mm3; time: 1min20s), 2) T2 mapping (17 echo multi shot TSE; ΔTE 7.6 ms; vox: 3x3x6mm3 with 6mm slice gap; time: 3min5s), and 3) Diffusion Tensor Imaging (Single shot SE-EPI, 42 diffusion weighted volumes; b-values from 1-600 mm/s²; vox: 3x3x6mm3; time: 3min30s).

Motor function assessment and muscle strength, using manual and quantitative testing, will be assessed in all participants.

Follow-up interval is 1 year for SMA patients and for controls between 12 and 18 years.

Structural and functional changes will be regarded in relation to clinical characteristics (e.g. duration of illness, disease progression), genetic factors and clinical scores.

Inclusion starts after approval of the local medical research ethics committee.

The MRI protocol has been developed and tested for calf muscles of adult volunteers.

We aim to investigate biomarker potential of various MRI techniques and to gain further insight into the pathogenic mechanisms of SMA.

63

Clinical importance of a single dystrophin gene mutation in women related to patients affected by Duchenne or Becker muscular dystrophy

Freja Fornander[1], BSc; Nanna Scharff Nielsen[1], BSc; Rasmus Møgelvang[2], MD, PhD; Anne-Sofie Vibaek Eisum[1], BSc; Annarita Ghosh Andersen[1], student; Morten Duno[3], PhD; Henning Bundgaard[2], PhD, John Vissing[1], MD, DMSc

[1]Copenhagen Neuromuscular Center, Department of Neurology, Rigshospitalet, University of Copenhagen, Denmark [2]Department of Cardiology, Rigshospitalet, Copenhagen, Denmark [3]Department of Clinical Genetics, Rigshospitalet, Copenhagen, Denmark [4]Department of Diagnostic Radiology, Neuro-orthopedic Radiology, Rigshospitalet, Copenhagen, Denmark

Single mutation carriers of recessively inherited myopathies are typically considered unaffected. Manifesting carriers, however, have been described in several myopathies, including Duchenne and Becker dystrophinopathies, but the frequency and extent of affection in female carriers have not been investigated prospectively.

Through an observational, prospective, cross-sectional study, female carriers of Duchenne and Becker muscular dystrophies are examined to understand the frequency with which carriers

Methods: Genetically verified female carriers of single DMD gene mutations are examined by MRI of the heart, lower back, thigh and calf muscles, isokinetic muscle dynamometry (thighs and calves), blood creatine kinase and heart muscle biomarkers levels, echocardiography and 24-48-hour Holter monitoring. Dixon MRI is used to establish muscle fat fractions of the lower extremity muscles and is correlated to muscle strength measurements. Furthermore, strength is also evaluated through the Medical Research Council (MRC) scale, and muscle pain and fatigue estimated by the Numerical Rating Scale and Fatigue Severity Scale.

The study is ongoing and final data are not yet available, but will be presented at the conference. However, so far, it is clear that 7 of 21 lower extremity scans to date are visually abnormal to some extent, in T1 sequences of thighs and calves. Predominantly affected muscles in these 7 women (mean age 52.5 [42; 60]) are hamstrings, the medial adductor muscles in the thigh (though not m. gracilis), and the gastrocnemii muscles - most being asymmetrically affected. Two of the females with visually abnormal scans are related to patients with a BMD phenotype, 4 with a DMD phenotype, and 1 is yet undefined. The mean age of the whole group (n=21) is 51.8 years [26; 67], with BMI average of 26.3. Self-reported average age of onset for symptoms (e.g muscle pain or loss of function – if present) was at 43.7 years of age (n=15).

These initial investigations point to affection of a third of carriers with DMD mutations, and that both carriers related to patients with DMD and BMD can be affected. Detailed data on muscle fat fraction levels and correlations with muscle strength testing, blood sampling and mutations type will be presented at the meeting.

64

Longitudinal diffusion-weighted whole-body MRI demonstrates dynamic changes in muscle integrity in motor neuron disease

IA Pierry, JJP Alix, DG Rao, N Hoggard, J Bigley, CJ McDermott, ID Wilkinson, PJ Shaw, TM Jenkins

Sheffield Institute for Translational Neuroscience, Academic Unit of Radiology, University of Sheffield, UK; Department of Neurosciences and Department of Neurophysiology, Royal Hallamshire Hospital, Sheffield, UK.

A major barrier to developing effective therapeutics in motor neuron disease (MND) is the lack of a biomarker to objectively track progression over short, inexpensive time-scales. Previous imaging studies have focused on the central nervous system.

To investigate the utility of a novel diffusion-weighted muscle MRI approach in MND. We hypothesised that whole-body methodology would capture inherent clinico-anatomical phenotypic heterogeneity, and facilitate a reductionist approach to analysis, allowi ng measurements from different body regions to be brought into a common domain.

Twenty-nine MND patients and 22 controls underwent whole-body diffusion-weighted MRI (3 Tesla, single-shot EPI, TR=9412ms, TE=66ms, TI=250ms,b=0,1000s/mm2voxelsize=2.3x2.3x5mm 3, 8 stations, apparent diffusion coefficient (ADC) axial acquisitions), at baseline, 4, and (in patients) 12 months. Mean ADC estimates were obtained from the tongue, thoracic paraspinals and bilateral tibialis anterior, and summed across body regions. Arm muscle analysis proved technically challenging and is ongoing. Clinical data and electrophysiological motor unit number index (MUNIX) estimates were collected. Unpaired and paired t-tests were used to assess between-group differences and longitudinal changes, respectively. Clinical, electrophysiological and radiological associations were assessed using linear regression.

MND patients exhibited lower multi-region ADC than controls at baseline (6055 vs 7395 mm^2/s, $p<0.001$), and patients' multi-region ADC increased over 12 months (+1515mm^2/s, $p<0.001$). There were no longitudinal changes in controls. Higher baseline ADC in right and left tibialis anterior was associated with greater clinical weakness in this muscle ($p=0.031$ and $p=0.004$, respectively), and lower MUNIX on the right ($p=0.001$). There were no associations between multi-region ADC and functional questionnaires, power scores or disease duration.

Dynamic changes in ADC were seen in MND patients. At baseline, lower ADC than controls reflects restricted diffusion, likely due to myofibrillar membrane damage and/or fat replacement. Conversely, greater clinical weakness and motor unit loss in MND patients was associated with higher baseline ADC in leg muscles likely due to active denervation resulting in intramuscular fluid shifts. Longitudinal ADC increases may reflect reduced cellularity due to clearance of cellular debris and tissue loss. Whole-body diffusion-weighted MRI appears to detect evolving pathophysiology in MND, offering potential as a novel biomarker.

65

Comparison of short and long diffusion times to assess muscle microstructure in patients with Becker Muscular Dystrophy

J. Burakiewicz[1], C. Baligand[1], M. T. Hooijmans[1], Z. Koeks[2], L. van der Weerd[1,3], E.H. Niks[2], J.J.G.M. Verschuuren[1], H.E. Kan[1]

[1]C.J. Gorter Center for High Field MRI, Department of Radiology [2]Department of Neurology [3]Department of Human Genetics, Leiden University Medical Centre, Leiden, The Netherlands

Diffusion Tensor Imaging (DTI) gives insight into tissue architecture at the cellular level by measuring microscopic molecule movement. Stimulated Echo (STE)-DTI allows to probe larger diffusion distances than the most commonly used spin echo approach by using longer diffusion times. This technique has not been applied in NMD patients before.

In this pilot study we evaluated the diffusion-time dependent behaviour of diffusion scalars in patients with Becker Muscular Dystrophy and healthy controls using STE-DTI.

Left calves of four patients with BMD and 5 healthy controls were scanned on a 3T Philips Ingenia system with fat-suppressed STE-DTI scans (diffusion times of 100ms and 300ms, TR/TE=5 s/56 ms, resolution 4x4x6 mm3 for BMD and 2x2x6 mm3 for healthy controls). Mean diffusivity (MD) and fractional anisotropy (FA) were calculated in six

muscles and compared on a muscle by muscle basis between short and long diffusion times using a paired t-test.

In BMD patients, FA values were similar for the lower and higher diffusion times (0.23 +/- 0.05 at 100ms versus 0.24 +/- 0.05 at 300ms, p=0.42), whereas in healthy controls, FA values increased significantly (0.23 +/- 0.03 versus 0.27 +/- 0.03, p=0.0043).

In BMD patients and healthy controls MD values were significantly lower at 300ms than at 100ms (1.31 +/- 0.09 versus 1.19 +/- 0.08 x10-3 mm2/s, p=0.0040 for BMD; 1.51 +/- 0.07 x10-3 mm2/s versus 1.39 +/- 0.08 x10-3 mm2/s, $p < 0.0001$ for controls).

Increase in FA and decrease in MD in healthy controls suggests that longer diffusion times allow us to probe barriers such as cell membranes with greater sensitivity. The lack of change in FA in BMD patients may be indicative of deterioration of these barriers, which is consistent with BMD studies showing increased membrane permeability due to the pathology. It should be noted that long diffusion times in healthy controls had signal-to-ratio close to the threshold below which FA is artificially lowered and MD raised.

By enabling longer diffusion times compared to conventional spin-echo DTI, STE-DTI may offer a new quantitative approach to muscle fibre alterations caused by Becker Muscular Dystrophy and other neuromuscular diseases.

66

Multi-centric evaluation of stability of quantitative outcome measures in healthy calf muscles

Lara Schlaffke[1,2,3], Robert Rehmann[2], Louise AM Otto[4], Jedrek Burakiewicz[3], Celine Baligand[3], Jithsa Monte[5], Chiel den Harder[5], Aart Nederveen[5], Hermien Kan[3], Martijn Froeling[1]

[1]Department of Radiology, University Medical Centre Utrecht, Utrecht, The Netherlands [2]Department of Neurology, BG-University Hospital Bergmannsheil, Ruhr-University, Bochum, Bochum, Germany [3]C.J., Gorter Center for High Field MRI, Department of Radiology, Leiden University Medical Centre, Leiden, The Netherlands [4]Brain Centre Rudolf Magnus, Department of Neurology, University Medical Centre Utrecht, Utrecht, The Netherlands [5]Department of Radiology, Academic Medical Center, Amsterdam, The Netherlands

Quantitative muscle MRI is increasingly used in the assessment of muscle status in neuromuscular diseases. Commonly used parameters include fat-fraction, T2-relaxation time and, more recently, diffusion properties. To include these parameters in clinical trials as outcome measures, high accuracy and reproducibility between different MR sites is vital. While physiological conditions, unrelated to diseases are known to affect these parameters, little is known about the temporal stability during rest.

This study aimes to i) investigate the stability of quantitative MRI parameters in in skeletal muscle in the usual time frame of an MR examination and ii) to evaluate the stability of commonly used techniques between MR-scanning sites.

QuantitativeT2, DTI and Dixon data were acquired on Philips 3T systems from the calf muscles of 22 healthy volunteers (11 males/11 females) in four different MR-centers (AMC Amsterdam 6, BMH Bochum 5, LUMC Leiden 4 and UMC Utrecht 7) and repeated six times within one hour. The T2 sequence contained 17 Echos (ΔTE=7.6ms) and was evaluated with the EPG method. For DTI 42 diffusion weighted gradients with b-values from 1-600mm/s² were acquired. The DTI fit (WLLS) was performed using all b-values as well as using only b-values >200mm/s² to correct for perfusion signal. Average values of all parameters for each muscle in the calf was calculated using manual segmentations based on a Dixon image.

Repeated measures ANOVA revealed significant location dependent decreases of the estimated T2 by ~1ms (UMC: -1.07ms, LUMC: -0.54ms, AMC: -1.02 ms, BMH: +0.18ms) which is not stabilizing within the observed timespan of 1hr. The SNR from the DWIs also decreased significantly (UMC: -1.79, LUMC: -1.21, AMC: -1.33, BMH: +0.01). The FA, which highly correlates to SNR ($p < .001$) also shows a significant decrease (-0.005), which stabilizes over time. It steadies after 30min and after only 10 min when eliminating the perfusion signal.

Evaluation of quantitative imaging outcomes dependents on the timepoint of the data acquisition within a 1 hour protocol. Different temporal dynamics were seen between scanners. Therefore, it is likely that observed changes were not only driven by physiological factors, like resting, but also by scanner stability and SNR.

67

Multimodal MR investigation of the lower leg muscles in patients with Becker muscular dystrophy

C. Baligand, M.T. Hooijmans , Z. Koeks, J. Burakiewicz J.J.G.M. Verschuuren, E.H. Niks, H.E. Kan

C.J. Gorter Center for High Field MRI, Department of Radiology, Department of Neurology, Department of Human Genetics, Leiden University Medical Centre, Leiden, The Netherlands

Quantitative MRI and MRS have been extensively used to study neuromuscular disorders. Parameters like fat fraction, T2 relaxation time, metabolite ratios and diffusion measures can serve as markers of disease progression. In Becker muscular dystrophy (BMD), dystrophin function is impaired due to a mutation in the DMD gene. This results in progressive fat infiltration and fibrosis, predominantly at later stages of the disease, while metabolic alterations such as changes in phosphodiester (PDE) levels occur prior to fat infiltration. Diffusion tensor imaging (DTI) can be sensitive to other structural changes such as membrane damage, fiber size, type, and content. Therefore, the relation between DTI metrics, fat infiltration and metabolic changes might be of value in the study of BMD.

Our goal was to examine the diffusion properties of the lower leg muscles in BMD muscles and their relation with differences in phosphodiester (PDE) and fat levels.

Eighteen BMD patients (age 44±14) and thirteen age-matched healthy subjects (Controls; age 40±15) were examined. During a single visit, we acquired: (a) fat fraction (FF) maps using three-point Dixon, (b) 31P 2D-Chemical Shift Imaging to assess PDE levels, and (c) spin-echo (SE)-DTI to assess diffusion properties. All imaging data were acquired at 3T, and 31P spectroscopy at 7T.

FF was significantly higher in BMD for all muscles compared to controls ($p < 0.001$), with a large range of values in BMD (4.1 to 51.7%). We found significantly higher PDE levels in BMD than in controls ($p=0.027$). Fractional anisotropy (e.g. BMD: 0.21±0.02, controls: 0.19±0.01 in soleus) and mean diffusivity (e.g. BMD: 1.66±0.06, CON: 1.65±0.03 10-3mm2.s-1 in soleus) did not show any differences between the two groups. No correlation was found between any of variables.

Similar to what we previously reported, PDE levels were high and independent of FF in BMD. SE-DTI parameters did not show differences compared to healthy controls and were not correlated to PDE or FF changes. It is possible that SE-DTI's sensitivity is limited due to short diffusion times. Future work will evaluate the use of longer diffusion times with a stimulated echo approach.

68

Quantification of rectus abdominis muscles in relation to functional tests in Becker Muscular Dystrophy

Naarding K.J.[1], Hooijmans M.T.[2], Koeks Z.[1], Baligand C.A.J.[2], Verschuuren J.J.[1], Kan H.E.[2], Niks E.H[1]

[1]Department of Neurology, Leiden University Medical Center, Leiden, The Netherlands [2]Department of Radiology, C.J. Gorter Center for High Field MRI, Leiden University Medical Center, Leiden, The Netherlands

Becker muscular dystrophy (BMD) is caused by mutations in the DMD gene leading to a truncated, partially functional dystrophin protein. Quantitative MRI (qMRI) is used to assess disease progression in muscular dystrophies by assessing the fat fraction (FF) and T2 relaxation time. Most studies have focussed on limb muscles, while truncal muscles are also important for balance and stability.

To assess the feasibility of quantifying rectus abdominis (RA) muscle FF in BMD patients and to study the relationship with functional tests.

Whole body 3D multi-station 2-point Dixon-gradient echo scans (FoV: 520x359x120mm3; voxel size 1.91x1.91x5.0mm3 per stack) were acquired at 3T in BMD patients and male healthy controls (HC). Regions of interest were drawn in the most distal slice in which the kidneys were still visible. FF was calculated as a weighted mean value of right and left RA. Functional tests performed in BMD patients included Vignos and North Star Ambulatory Assessment (NSAA). Differences in FF were assessed with a Mann–Whitney U test.

RA images were obtained in 16 ambulant BMD patients (19-66 years) and 8 HC (28-48 years). Median FF was significantly higher in BMD compared to HC (16.5%, range 7.9-50.2% versus 10.9%, range 7.3-16.9%, $p=0.009$). Median FF of 10

BMD patients with a Vignos score ≤3 was slightly below that of 6 patients with a Vignos score ≥4 (16.5%, range 7.9-34.5% versus 19.5%, range 12.0-50.2%), although this was not statistically significant (p=0.562). NSAA sit-up sub-score was available for 13 BMD patients. Eleven patients had a maximum score of 2 and showed a wide range of FF (7.9-30.8%). Only 2 patients had a sub-score of 1 and had a FF of 16.9% and 50.2% respectively.

Quantification of rectus abdominis muscle fat fraction in BMD patients is feasible and elevated compared to healthy controls. We could not find a relation with clinical parameters, probably due to the small sample size, and the fact that all BMD patients were still ambulant and thus relatively mildly affected.

69

Specific strength is reduced in facioscapulohumeral dystrophy muscles. An MRI-based musculoskeletal analysis

Linda Heskamp[1], Marco Marra[2], Karlien Mul[3], Baziel van Engelen[3], Nico Verdonschot[2], Arend Heerschap[1]

[1]Radboud university medical center, Radiology and Nucleair Medicine, Nijmegen, The Netherlands [2]Radboud university medical center, Orthopaedic Research Laboratory, Nijmegen, The Netherlands [3]Radboud university medical center, Neurology, Nijmegen, The Netherlands

Patients with facioscapulohumeral dystrophy (FSHD) experience muscle weakness, which is associated with fat infiltration and atrophy. In addition, in other muscular dystrophies, Duchenne and Becker muscular dystrophy, it has been shown that the specific muscle strength, i.e. the strength per unit area of actual muscle tissue, is also reduced and thus it represents an additional factor for muscle weakness. Whether this is also the case in FSHD, is still not known.

Our aim was to asses if specific muscle strength is reduced in FSHD patients, as compared with healthy controls.

Ten FSHD patients and ten healthy volunteers were retrospectively included. MR images and maximum voluntary isometric contraction (MVIC) torques of the quadriceps muscles were acquired for all subjects. Muscles contours were manually delineated on 2pt Dixon MR images to calculate anatomical volumes of each muscle. Fat fractions in muscles of FSHD patients were derived from the 2pt DIXON to correct for non-contractile muscle volume. Thereafter, the muscle strength and physiological cross-sectional area (PCSA) were determined using a musculoskeletal model. PCSA is calculated as contractile muscle volume divided by the optimal fiber length and corrected for the pennation angle. Finally, specific strength was calculated for each subject as muscle strength divided by the total PCSA. The difference in MVIC, PCSA and specific strength between FSHD and control groups was tested for significance with an unpaired t-test.

Compared to healthy controls FSHD patients have a significantly lower MVIC (208.3±23.2 Nm vs 63.1±12.1 Nm, p<0.0001) and PCSA (90.5±11.9 vs 145.1±15.7, p=0.0125). Specific strength of quadriceps muscles was significantly reduced in FSHD patients compared to healthy controls (19.7±1.9 N/cm2 vs 44.4±2.4 N/cm2, p<0.0001).

FSHD patients generate less force per unit quadriceps muscle PCSA than healthy controls, even when correcting for fat infiltration and atrophy. Possible explanations are a reduction in the intrinsic force generating capacity of muscle fibers in FSHD, impaired force propagation due to fatty infiltration and/or conversion of strong fast-twitch fibers to weaker slow-twitch fibers.

70

Quantitative MRI in Myotonic Dystrophy Type 1: Natural progression and correlation with functionality

Linda Heskamp[1], Marlies van Nimwegen[2], Guillaume Bassez[3], Cecilia Jimenez-Moreno[4], Marieke Ploegmakers[1], Jean-Francois Deux[5], Grainne Gorman[6], Hanns Lochmuller[4], Michael Catt[7], Baziel van Engelen[2], Arend Heerschap[1]

[1]Radboud university medical center, department of Radiology and Nuclear medicine, Nijmegen, The Netherlands [2]Radboud university medical center, department of Neurology, Nijmegen, The Netherlands [3]Henri Mondor University Hospital Neuromuscular Reference Center, Paris, France [4]Newcastle University Institute of Genetic Medicine Newcastle UK [5]Henri Mondor university Hospital Radiology Paris France [6]Newcastle University Institute for Ageing and Health Newcastle UK [7]Newcastle University Institute of Neuroscience Newcastle UK

Quantitative MRI (qMRI) is a valuable tool for the non-invasive assessment of diseased muscles.

We used qMRI to investigate the occurrence and progression of fatty infiltration, atrophy and edema-like processes in muscles of Myotonic Dystrophy Type 1 (DM1) patients and correlated this with their functionality.

At two sites, Nijmegen and Paris, we included 33 DM1 patients and 10 healthy controls. All subjects underwent an MRI of one of their legs. Muscles contours of 12 thigh and 8 calf muscles were manually delineated on the 2pt Dixon MR images to determine the fat percentage (FF) and muscle volume (MV). Furthermore, T2 relaxation time of muscle water (T2m) was determined by voxel-wise fitting multi-spin echo images using an extended phase graph model. Thirteen patients underwent a follow-up MR scan at 10 months. Functionality was tested with a 6 minute walking test (6MWT) and muscle impairment rating score (MIRS). In 21 DM patients daily life activity was assessed for two weeks with an actometer, that recorded the average activity over 24 hours, the most active and the least active 5 hours.

Compared to healthy controls DM1 muscles showed significant ($p<0.05$) increased average FF (3.7%±1.5% vs 15.6%±11.1%) and T2m (32.3ms±0.7ms vs 33.7ms±2.2ms) and decreased MV (1089cm3±246cm3 vs 903cm3±233cm3). The calf muscles were more affected than the thigh muscles, with the gastrocnemius medialis and soleus being predominantly affected in the calf. After 10 months, FF increased significantly in DM1 (1.22±1.2%, $p<0.05$), while MV remained stable. The 6MWT and MIRS correlate significantly with FF ($p<0.01$; Pearson's r=-0.56 and $p < 0.01$; Spearman′s rho= 0.54). Also, average activity over 24 hours and 5 most active hours correlate with FF ($p<0.001$; Pearson r =-0.80 and r=-0.79, respectively), while no correlation is found with the 5 least active hours.

Muscles in DM1 are affected in a specific pattern and show a slow, but significant progression of fat infiltration over 10 months. The significant correlation of FF with 6MWT and MIRS indicates that severity of fatty infiltration greatly impacts functionality. Interestingly, this qMRI study shows that activity of the patients in daily life is associated with their muscular FF.

71

Muscle functional oxidative capacity varies along the length of healthy human tibialis anterior

Linda Heskamp, Andreas Boss, Vincent Breukels, Lauren J Bains, Mark J van Uden, Arend Heerschap

Department of Radiology and Nuclear Medicine, Radboud university medical center, Nijmegen, The Netherlands

The rate of phosphocreatine (PCr) recovery (kPCr), characterizing muscle oxidative capacity, is traditionally assessed with unlocalized 31P magnetic resonance spectroscopy (MRS) using a single surface-coil, although skeletal muscle is known to be non-uniform. For example, in rodents fiber distribution was shown to vary along the length of the tibialis anterior (TA). The proportion of oxidative fibers is higher in the proximal part of the muscle, whereas glycolytic fibers are more prominent distally. Whether this applies to humans, is still unknown. Oxidative type I fibers have a higher capacity for PCr-recovery as compared to glycolytic type II fibers, which might result in a higher kPCR in the proximal part of the TA muscle compared to distally.

The aim of our study is to investigate if kPCr varied along the length of the TA muscle in ten healthy male volunteers.

A commercial 31P/1H-volume coil for transmit was combined with a home-built 31P phased-array for receive, optimized for TA. Localization was achieved by the limited sensitivity profile of the five coil elements and their position along the muscle. Mono-exponential kPCr was determined from each of the five elements after 40 seconds of submaximal isometric dorsiflexion (SUBMAX) or an incremental exercise to exhaustion (EXH) inside a 3T MR-system. In addition, muscle functional MRI (1H-mfMRI) was performed during and after another 40 seconds of submaximal contractions.

A strong gradient in kPCr was observed along TA. kPCR was two times higher proximally vs. distally (SUBMAX: 0.94 min-1 distally vs 2.05 min-1 proximally; EXH: 0.61 min-1 distally vs 1.75 min-1 proximally; $p<0.001$). Statistical analysis showed that pH cannot explain this gradient. Also the relative increase of 1H-mfMRI signal and slope of the initial post-exercise signal change was higher proximally (p between 0.001 and 0.004) and strongly correlated with kPCr.

Pronounced differences in the functional oxidative capacity along human TA was measured with 31P MRS and 1H-mfMRI. This unique finding may reflect non-uniform fiber type distribution or capillary density and regional variation in the metabolic or biomechanical demands of everyday activities on this muscle.

72

Contractile cross-sectional area versus muscle strength in patients with congenital and RYR1 myopathies

Anne-Sofie Vibæk Eisum, BSc Med, Tove Maria Freja Fornander, BSc Med, Nanna Scharff Nielsen, BSc Med, Annarita Ghosh Andersen, medical student, Julia R. Dahlqvist, MD, Nanna Witting MD, PhD, John Vissing, MD, DMSci

Copenhagen Neuromuscular Center, Department of Neurology, Rigshospitalet, University of Copenhagen, Denmark

Congenital myopathy (CM) patients are characterized by hypotonia at birth. Subtypes are defined by muscle histology and genetic etiology. Mutations are often found in genes encoding proteins involved in the sarcomere protein structure. RYR1-myopathy (RYR1) is caused by mutations in the RYR1-gene, important in regulating the calcium flow involved in sarcomere function. Mutations can lead to various myopathies, among those CM and adult onset rhabdomyolysis-myalgia syndrome. Both patient groups have afflicted muscles on sarcomeric level, which potentially could compromise contractility.

The aim of the study is to investigate whether CM and RYR1 patients have a disrupted relationship between muscle strength and the contractile cross-sectional area (CCSA), which is the fat-free cross-sectional area of the muscle. In healthy individuals, there is a close direct correlation between strength and CCSA.

Maximal isokinetic muscle strength is investigated by an isokinetic dynamometer, measuring Peak Torque (PTq). Dixon MRI technique quantifies muscle fat fraction of thigh and calf muscles, from which CCSA can be calculated.

Preliminary data shows that strength-CCSA relationship for the most part is preserved in RYR1 (n=6) and CM (n=6) myopathies, but in some the relationship seems disrupted.

The findings indicate a perturbed contractile function in some CM patients. Further studies will unravel in which genetic subtypes this sarcomeric dysfunction occurs.

73

Inflammation and fat replacement of muscle in patients with FSHD

Julia R. Dahlqvist, MD; Nanna S. Nielsen, BSc; Sofie T. Østergaard, BSc; Freja Fornander, BSc; Anne-Sofie V. Eisum, BSc; Carsten Thomsen, MD, PhD; John Vissing, MD, PhD

[1]Copenhagen Neuromuscular Center, Department of Neurology, Rigshospitalet, University of Copenhagen, Denmark [2]Department of Diagnostic Radiology, Rigshospitalet, Denmark

Facioscapulohumeral dystrophy (FSHD) is the second most common autosomal dominant muscular dystrophy, and is characterized by progressive asymmetric weakness and atrophy of facial, scapular and proximal arm muscles. The clinical presentation varies, and leg, abdominal and back muscle involvement is also common. Muscle affection is often asymmetric and progression is often stepwise and not continuous as in most other muscular dystrophies. It has been suggested that muscle inflammation contributes to the pathophysiology of FSHD and that it predates the destruction of muscle and its conversion to fat tissue. However, the duration of inflammatory lesions in muscle and whether they always progress to destruction of muscle in FSHD is unknown.

In this study, we aim to shed light on the pathophysiology of FSHD by following inflammatory lesions with sequential MRI over 2 years in patients with FSHD. The questions we ask are whether healthy muscle can progress to fatty degeneration without inflammation, how long time the inflammatory lesions last, and whether inflammation can be resolved without degeneration of muscle or are invariably followed by fat degeneration of the muscle.

FSHD patients with a molecular diagnosis of FSHD type 1 are followed over 2 years with sequential evaluations every 2nd (year 1) to 4th (year 2) month.

Short tau-inversion recovery sequences (STIR) and T2-mapping are used to detect and quantify the degree of muscle inflammation in the legs. The MR

Dixon technique is utilized to perform muscle fat fraction analysis of the leg muscles. Muscle strength in legs are assessed with a Biodex dynamometer. All subjects answer questions on pain at every visit and blood samples are drawn and analyzed for muscle damage and inflammatory markers.

Ten FSHD patients with a molecular diagnosis of FSHD type 1 (28-62 years) have been included. They have been followed over approximately 1 year. Preliminary MRI results suggest that inflammation is a long-lasting process with no visually detectible changes in fat content after only one year.

This is an ongoing study and we have not analyzed any data yet. We will have some results by November 2017.

74

Muscle involvement in patients with SBMA

Julia R. Dahlqvist, MD; Sofie T. Østergaard, BSc; Nanna S. Nielsen, BSc; Freja Fornander, BSc; Carsten Thomsen, MD, PhD; John Vissing, MD, PhD

[1]Copenhagen Neuromuscular Center, Department of Neurology, Rigshospitalet, University of Copenhagen, Denmark [2]Department of Diagnostic Radiology, Rigshospitalet, Denmark

Spinal and bulbar muscular atrophy (SBMA), also called Kennedy disease, is an X-linked disease caused by a CAG-repeat expansion in the gene encoding the androgen receptor (AR). The motor dysfunction in SBMA is probably caused by a toxic effect of the mutant AR on both neurons and skeletal muscle. SBMA is slowly progressive with weakness and atrophy of the bulbar and extremity muscles. However, the skeletal muscle involvement has not been investigated systemically in this patient group.

In this study, we aim to investigate which skeletal muscles are affected in SBMA.

Patients with a molecular diagnosis of SBMA were included. The Dixon technique of MRI was utilized to perform muscle fat fraction analysis of the arm, back and leg muscles. Muscle strength in arms and legs were assessed with a Biodex dynamometer.

Thirty-nine patients with SBMA have been included and 37 have been examined so far. The data will be analyzed during Summer 2017 and we will be able to present the results at the conference.

The results have not been analyzed yet.

75

Adding quantitative muscle MRI to the FSHD clinical trial toolbox

Karlien Mul MD[1]; Sanne C.C. Vincenten[1]; Nicol C. Voermans MD, PhD[1]; Richard J.L.F. Lemmers PhD[2]; Patrick J. van der Vliet BSc[2]; Silvère M. van der Maarel PhD[2]; George W. Padberg MD, PhD[1]; Corinne G.C. Horlings MD, PhD[1]; Baziel G.M. van Engelen MD, PhD[1]

[1]Department of Neurology, Radboud University Medical Center, Nijmegen, the Netherlands [2]Department of Human Genetics, Leiden University Medical Center, Leiden, the Netherlands

Since the discovery of the (epi)genetic origin of FSHD, new therapeutic strategies are being developed and clinical trials are at the near horizon. Consequently, there is an urgent need for a 'clinical trial toolbox', including biomarkers to detect subtle changes in muscle pathology. Quantitative muscle MRI may provide such a biomarker, however evidence of its correlation to clinical outcome measures still has to be established.

Adding quantitative muscle MRI to the clinical trial toolbox for FSHD by correlating it to clinical outcome measures in a large cohort of genetically and clinically well-characterized FSHD patients comprising the entire clinical spectrum.

Quantitative MRI scans of leg muscles of 140 FSHD1 and FSHD2 patients were assessed for fatty infiltration and TIRM hyperintensities and were correlated to multiple clinical outcome measures.

The mean fat fraction of the total leg musculature correlated highly with the Motor Function Measure, FSHD clinical score, Ricci score and 6-minute walking test (correlation coefficients -0.845; 0.835; 0.791; -0.701 respectively). Fat fraction per muscle group correlated well with corresponding muscle strength (correlation coefficients up to -0.82). The hamstring muscles, adductor muscles, m. rectus femoris and the m. gastrocnemius medialis were affected most frequently, also in early stage disease and in patients without leg muscle weakness. Muscle involvement was asymmetrical in 20% of all muscle pairs and fatty infiltration within muscles showed a decrease from distal to proximal of 3.9%. TIRM hyperintense areas, suggesting inflammation, were found in 3.5% of all muscles, with and without fatty infiltration.

We show a strong correlation between quantitative muscle MRI and clinical outcome measures.

Muscle MRI is able to detect muscle pathology before clinical involvement of the leg muscles. This indicates that quantitative leg muscle MRI is a promising biomarker that captures disease severity and motor functioning and can thus be included in the 'FSHD trial toolbox'.

76

Spatially localized phosphorous metabolism of skeletal muscle in Duchenne Muscular Dystrophy patients: 24 –month follow-up

M.T. Hooijmans[1], N. Doorenweerd[1,3], C. Baligand[1], J.J.G.M. Verschuuren[2], I. Ronen[1], E.H. Niks[2], A.G. Webb[1], H.E. Kan[1]

[1]Dept of Radiology, C.J. Gorter Center for High Field MRI, Leiden University Medical Center, Leiden, The Netherlands [2]Dept of Neurology, Leiden University Medical Center, Leiden, The Netherlands [3]John Walton Muscular Dystrophy Research Centre, Newcastle University, Newcastle Upon Tyne, United Kingdom

Quantitative MR of muscle is increasingly important as potential outcome measure for therapy development in Duchenne Muscular Dystrophy (DMD). Since therapy is aimed at preserving or improving muscle tissue, a marker should reflect changes within the muscle and should have sufficient discriminative power. Unfortunately, neither %fat nor water T2 fulfill these criteria.

To assess changes in phosphodiester (PDE)-levels, detected by spatially resolved 31P MRS, over 24-months to determine the potential of PDE as marker for muscle tissue changes in DMD patients.

31P 2D-CSI datasets were acquired in the right lower leg of 18 DMD patients (range: 5-15.4 years) and 12 age-matched controls (range: 5-14 years) at three time-points (baseline, 12-months, and 24-months) using a 7T MR-System (Philips Achieva). 3-point Dixon images were acquired at 3T (Philips Ingenia) to determine muscle fat fraction. Analyses were done for six muscles that represent different stages of muscle wasting. Differences between groups and time-points were assessed with non-parametric tests with correction for multiple comparisons. Coefficient of variance (CV) were determined for PDE in four healthy adult volunteers in high and low signal-to-noise ratio (SNR) datasets.

PDE-levels were significantly higher (two-fold) in DMD patients compared to controls in all analyzed muscles at almost every time point and did not change over the study period. Fat fraction was significantly elevated in all muscles at all time points compared to controls, and increased significantly over time, except in the tibialis posterior muscle. The mean within-subject CV for PDE-levels was 4.3% in datasets with high SNR (>10:1) and 5.7% in datasets with low SNR.

The stable two-fold increase in PDE-levels found in DMD patients in muscles with different levels of muscle wasting over 2-year time, including DMD patients as young as 5.5 years-old, suggests that PDE-levels may increase very rapidly early in the disease process and remain elevated thereafter. The low CV values in high and low SNR datasets show that PDE-levels can be accurately and reproducibly quantified in all conditions. Our data confirms the great potential of PDE as a marker for muscle tissue changes in DMD patients.

77

Muscle contractility of calf and thigh muscles in patients with mitochondrial myopathies

Nanna Scharff Poulsen, BSc[1], Julia Dahlqvist, MD[1] Kirsten Lykke Knak, PT, MSc[1], Nicoline Løkken, MD[1], Gitte Hedermann, MD[1] and John Vissing, MD, DMSc[1]

[1]Copenhagen Neuromuscular Center, Rigshospitalet, Copenhagen University Hospital, Copenhagen, Denmark

A hallmark of healthy muscles is a linear relation between muscle strength and muscle cross sectional area (CSA). In patients with mitochondrial myopathies (MM), cellular energy metabolism is compromised, influencing especially skeletal muscles, thereby reducing the force generated after many contractions. Furthermore, structural muscle changes can be present, such as replacement of muscle by fat. Mice studies have indicated that perturbed mitochondrial metabolism may even affect contractility of initial muscle contractions.

The aim is to investigate the contractility of calf and thigh muscles in patients with MM.

The maximal strength of 36 subjects with MM and 38 healthy controls was investigated using a Biodex dynamometer. CSA was measured using MRI. Through Dixon sequences, fat fractions of muscles

were calculated, and by subtracting fat areas from the CSA, the contractile CSA (CCSA) was found. The contractility of muscles was investigated by comparing strength with CCSA.

Strength, CSA and CCSA were significantly reduced in patients with MM compared to healthy controls. Fat fractions were significantly increased. The strength/CCSA relationship, however, was not affected, and likewise, the slope of the linear regression line of strength and CCSA in patients with MM was to that of healthy controls.

Our findings suggest normal contractile properties of initial contractions in thigh and calf muscles of patients with MM, even though strength, CSA, and CCSA were all lower than normal.

78

An accurate supervised segmentation of 3D individual lower leg muscles from 7T-MRI using label propagation

A. Ogier[1], A. Fouré[1], M. Sdika[2], A. Le Troter[1], D. Bendahan[1]

[1]Aix Marseille Univ, CNRS, CRMBM, Marseille, France
[2]Univ.Lyon, INSA-Lyon, Université Claude Bernard Lyon 1, UJM-Saint Etienne, CNRS, Inserm, CREATIS UMR 5220, LYON, France

Manual and automated segmentation of individual muscles in magnetic resonance images have been recog-nized as challenging given the high variability of shapes between muscles and subjects and the discontinui-ty or lack of visible boundaries between muscles. So far, manual segmentation of anatomical structures has been used in multiple studies and this approach has been widely acknowledged as time-consuming and operator-dependent.

In the present study, we proposed an original algorithm allowing a 3D semi-automatic segmentation of indi-vidual muscles from MR images based on the transversal propagation of manually-drawn 2D masks.

Our method is semi-automatic since it requires the manual selection of initial seed regions (composed of a set of 2D labeled masks) corresponding to several slices within the region of interest (3D). It corresponds to the fusion of both propagation and interpolation methods based on several ascending and descending non-linear registrations. The aim is to generate the labels of each individual muscle on the overall dataset from the propagation of the manually provided anatomical information in a few slices within the region of interest. The propagation strategy is based on successive registrations of each grey level from one slice to another in order to follow the local deformations of the muscles. The interpolation strategy is based on registrations of manual masks for the whole set of slices located between slices manually-segmented. Our fusion ap-proach was evaluated on a database consisting of 19 sets of lower leg MR images of healthy volunteers scanned at 7T. T1-weighted images (200 slices, field of view = 180 × 180 mm; matrix = 384 × 384; slice thickness = 1.5 mm; no gap between slices; acquisition time = 8 min 34 s) were recorded using a gradient recalled echo pulse sequence. The seed regions we used were the manually segmented slices bordering the region of interest and four additional slices located at the borders of lower leg muscles appearing or disappearing along the leg. The DICE similarity coefficient (DSC) and the muscle volume similarity fraction (MVSF) were used to estimate the performance of our method with respect to a manual segmentation of all the muscles of the lower leg.

linear registrations. The aim is to generate the labels of each individual muscle on the overall dataset from the propagation of the manually provided anatomical information in a few slices within the region of interest. The propagation strategy is based on successive registrations of each grey level from one slice to another in order to follow the local deformations of the muscles. The interpolation strategy is based on registrations of manual masks for the whole set of slices located between slices manually-segmented. Our fusion ap-proach was evaluated on a database consisting of 19 sets of lower leg MR images of healthy volunteers scanned at 7T. T1-weighted images (200 slices, field of view = 180 × 180 mm; matrix = 384 × 384; slice thickness = 1.5 mm; no gap between slices; acquisition time = 8 min 34 s) were recorded using a gradient recalled echo pulse sequence. The seed regions we used were the manually segmented slices bordering the region of interest and four additional slices located at the borders of lower leg muscles appearing or disap-pearing along the leg. The DICE similarity coefficient (DSC) and the muscle volume similarity fraction (MVSF) were used to estimate the performance of our method with respect to a manual segmentation of all the muscles of the lower leg.

Results: Using only six slices manually-segmented as seed regions for the propagation to the 184 re-

main-ing slices, we obtained average DSC values ranging from 0.86 to 0.97 and MVSF values ranging from 0.01 to 0.09. DICE values higher than 0.90 were obtained for almost all the muscles (GM, SL, SO, FHL, TP, LCLL and ACLL) and the corresponding MVSF values were lower than 0.07. For the FDL muscle, the DICE and MVSF values were 0.86 and 0.09 respectively.

We have presented a semi-automated method allowing a segmentation of each lower leg muscle from MR images. The method was based on an automatic transversal propagation of manually-drawn masks based on several 2D non-linear registration approaches. We mainly showed that our propagated segmentation was very accurate with an averaged DSC value higher than 0.94 and robust for a minimal input of manually-segmented slices (3% of the total slice number).

79

Long-term follow-up of MRI changes in thigh muscles of patients with Facioscapulohumeral dystrophy: a quantitative study

Emmanuelle Salort-Campana[1,3,4], Farzad Fatehi[1,2,5], Arnaud Le Troter[2], Emilie Lareau-Trudel[1,2,6], Mark Bydder[2], Alexandre Fouré[2], Maxime Guye[2], David Bendahan[2], Shahram Attarian[1,3,4]

[1]Centre de référence des maladies neuromusculaires et de la SLA, Centre hospitalier universitaire la Timone, Université Aix-Marseille, Marseille, France [2]Aix-Marseille Université, Centre de Résonance Magnétique Biologique et Médicale, UMR CNRS 7339, Marseille, France [3]Aix Marseille University, INSERM, GMGF, Marseille, France [4]FILNEMUS [5]Iranian Center of Neurological Research, Shariati Hospital, Tehran University of Medical Sciences, Tehran, Iran [6]Département de médecine, division neurologie, Université de Sherbrooke, Sherbrooke, Quebec, Canada

Facioscapulohumeral muscular dystrophy (FSHD) is one of the most common hereditary muscular disorders. Currently FSHD has no known effective treatment and detailed data on the natural history are lacking. Determination of the efficacy of a given therapeutic approach might be difficult in FSHD given the slow and highly variable disease progression. Magnetic resonance imaging (MRI) has been widely used to qualitatively and quantitatively evaluate in vivo the muscle alterations in various neuromuscular disorders.

The main aim of the present study was to investigate longitudinally the time-dependent changes occurring in thigh muscles of FSHD patients using quantitative MRI and to assess the potential relationships with the clinical findings.

Thirty-five FSHD1 patients (17 females) were enrolled. Clinical assessment tools including manual muscle testing using medical research council score (MRC), and motor function measure (MFM) were recorded each year for a period ranging from 1 to 2 years. For the MRI measurements, we used a new quantitative index i.e. the mean pixel intensity (MPI) calculated from the pixel-intensity distribution in T1 weighted images. The corresponding MPI scores were calculated for each thigh, for each compartment and for both thighs totally (MPItotal). The total mean pixel intensity (MPItotal) refers to the sum of each pixel signal intensity divided by the corresponding number of pixels. An increased MPItotal indicates both a raised fat infiltration together with a reduced muscle volume thereby illustrating disease progression.

Clinical scores did not change significantly over time whereas MPItotal increased significantly from an initial averaged value of 39.6 to 41.1 with a corresponding rate of 0.62/year. While clinical scores and MPItotal measured at the start of study were significantly related, no correlation was found between the rate of MPItotal and MRC sum score changes, MFMtotal and MFM subscores. The relative rate of MPItotal change was 2.3% (0.5 – 4.3)/year and was significantly higher than the corresponding rates measured for MRCS 0% (0 – 1.7) /year and MFMtotal 0% (0 – 2.0) /year ($p = 0.000$).

On the basis of these results, we suggested that muscle MRI and more particularly the MPItotal index could be used as a reliable biomarker and outcome measure of disease progression. In slowly progressive myopathies such as FSHD, the MPItotal index might reveal subclinical changes, which could not be evidenced using clinical scales over a short period of time.

80

Combined quantification of fatty infiltration, T1- and T2*-relaxation times in normal-appearing skeletal muscle of controls and dystrophic patients

B. Leporq[1], A. Le Troter[2], Y. Le Fur[2], E. Salort-Campana[3], M. Guye[2], O. Beuf[1], S. Attarian[3], D. Bendahan[2]

[1]Université de Lyon, CREATIS; CNRS UMR 5220; Inserm U1206; INSA-Lyon; Université Lyon 1; Villeurbanne, France [2]Aix-Marseille University, CRMBM, CNRS UMR 6612, Marseille, France [3]Reference Center for Neuromuscular Disorders, Timone Hospital, Marseille, France

Over the last decades, MRI has emerged as a potential tool of interest in order to monitor tissue changes occurring in dystrophies. Although quantitative methods have been recently used, they have been generating a single MRI index at a time and the corresponding segmentation process, if any, has been performed manually.

To evaluate the combination of a fat-water separation method with an automated segmentation algorithm as a mean of quantifying tissue alterations in dystrophic patients.

Seven patients with a facio-scapulo-humeral dystrophy and 8 controls agreed to participate in the study after an informed consent has been obtained. Disease severity was assessed using commonly-used scales for the upper and lower limbs. MR acquisitions were performed at 1.5T using a 3D FLASH sequence repeated with 3 different flip angle values (5, 10 and 15°), 8 echo times (from 2.38 to 19.04 ms), TR = 22 ms, 1.7mm2 in plane resolution, 4 mm thickness.

Fat-water separation and T1 mapping : The fat-water separation method uses a specific phase correction process addressing the issues of wrapping, zero order phase errors linked to RF penetration and eddy current effect, and first-order phase errors linked to local B0 inhomogeneities in order to reconstruct the off-resonance demodulated real part images. We generated proton density fat fraction maps (PDFF) and relaxation times maps (T2* and T1).

Segmentation algorithm: The segmentation algorithm distinguished adipose tissue and normal-appearing muscle from the T2* map and combined active contours, a clustering analysis and a morphological closing process to calculate the index of fatty infiltration (IFI) in the muscle compartment. IFI was defined as the relative amount of fat pixels in the normal appearing muscle volume.

In patients, relaxation times were longer and fatty infiltration in the normal-appearing muscle was larger while T2* and PDFF distributions were broader. The relaxation times were correlated to the Vignos scale whereas the microscopic fatty infiltration was linked to the Medwin-Gardner-Walton scale. The IFI was linked to a composite clinical severity scale gathering the whole set of scales. We quantified MRI indices within the normal-appearing muscle volume using a conventional FLASH sequence including 8 echo times combined to a segmentation pipeline. Interestingly, these MRI indices could be considered as biomarkers of interest in order to monitor tissue changes occurring in dystrophies.

Journal of Neuromuscular Diseases 4 (2017) S59–S63
IOS Press

Author Index

ISSN 2214-3599/17/$35.00