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Workshop report

247th ENMC International Workshop: Muscle magnetic resonance imaging - Implementing muscle MRI as a diagnostic tool for rare genetic myopathy cohorts. Hoofddorp, The Netherlands, September 2019

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1. Introduction

On September 2019, 19 participants from 9 countries attended the 247th European Neuromuscular Center (ENMC) workshop in Hoofddorp, The Netherlands, to discuss the standardization of muscle magnetic resonance (MR) imaging protocols, the development of a muscle MR imaging databank, and increasing the education and training of key MR imaging features of genetic myopathies for the wider medical community. The group included researchers and clinical experts in neuromuscular disease (NMD) MR imaging, as well as a young researcher and a patient representative from the European Patients Forum (EPF).

Dr. Alexandra Breukel, Managing Director, ENMC, welcomed the participants and outlined the successful outcomes of the ENMC workshops over the past 25 years, including improving diagnostic tools, refining clinical trial design and importantly, increasing collaborations between researchers and scientists [1]. Dr. Breukel also highlighted the importance of patient participation as key stakeholders in clinical research including outcome measures, patient registries and biobanks [2]. Dr. Breukel thanked the boards of patient organizations and the pharmaceutical sponsors for their support.

1.1. Importance of imaging for patients with genetic myopathies

Genetic myopathies are a heterogenous group of hundreds of different diseases. Many subgroups such as muscular dystrophies and congenital myopathies are characterized by weakness and muscle degeneration leading to significant physical disability [3-5]. Patients with these disorders demonstrate a broad spectrum of overlapping phenotypes, with skeletal muscle deterioration ranging from mild to severe, and in many cases, a reduced life expectancy. Advances in diagnostic genetic technologies have shown that currently there are > 500 genetically distinct NMDs, half of them genetic myopathies, many of which share similar clinical features (www.musclegenetable.fr) [6].

Although molecular diagnosis through genetic testing remains the gold standard, a proportion of patients with rare genetic myopathies remain undiagnosed due to either lack of a plausible candidate gene or too many variants in a number of different genes [3,7,8]. Muscle biopsies suggest a diagnosis in a number of patients and can facilitate interpretation of genetic testing. However, they are invasive and expensive

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and many genetic myopathies are not identifiable on muscle biopsy due to unspecific findings or potential false negative sampling. Therefore, there is an unmet need for additional techniques to improve the diagnostic rate in these patients.

Muscle MRI shows the distribution and the extent of replacement of muscle tissue by fat, edema, and atrophy [9] and reveals the evolution of this pattern of muscle pathology over time [10-12]. The most commonly utilized diagnostic muscle MRI Sequences include T1-weighted [T1) Turbo Spin-Echo (TSE) (fat sensitive) and T2-weighted (T2) sequences with fat suppression by inversion recovery, which are T2-STIR (short tau inversion recovery) (fluid sensitive) or Turbo inversion recovery magnitude (TIRM). MRI can be used to characterize the distribution of both affected and spared muscles to define a specific, and sometimes pathognomonic, pattern of involvement [9,10,13-17]. MRI can also be applied to quantify the progression of muscle pathology longitudinally in a non-invasive fashion, by assing the fat fraction, relaxation time mapping or other techniques [18,19]. Currently, disease progression for genetic muscle diseases is documented by clinical assessment and functional scales, which are dependent on patient cooperation with often poor inter-rater reliability. Importantly, quantitative MRI is now also more frequently used as an outcome measure in clinical trials as it can sensitively monitor response to treatment [19-21] and even predict clinical deterioration [22,23]. Conventional MRI can provide important information about appropriate regions of interest to be targeted by quantitative imaging for clinical trials. MRI can also identify relevant targets for muscle biopsy that could not be easily identified clinically and subsequently can decrease the risk of a ‘negative’ or futile biopsy [24,25].

An increasing number of studies are using imaging in patients with genetic myopathies, however, many limitations have affected the implementation of MRI as a standard diagnostic tool [26]. Disease rarity has limited progress in determining imaging patterns in many genetic myopathies because 1) the majority of studies have low numbers of participants/scans and a mechanism to systematically facilitate sharing of anonymized images to develop larger cohorts is currently lacking [27]; 2) the proficiency to interpret MRIs is limited to a few centers with larger patient cohorts and an established interest in muscle imaging; 3) the variations in imaging protocols used in different centers make combining results difficult to achieve [26]. Many of the studies published so far include patients in mid-stages of their disease, less frequently patients in early or late stages. More natural history studies are required to follow disease progression in order to understand the full range of potential combinations than can be found in a single disease. There is an urgent need to standardize best practice guidelines to acquire, collect and assess MR images, as these data will help to define selective patterns and spectra of pathology, will contribute to our understanding of pathomechanisms of disease and possibly identify the most relevant regions of interest for quantitative MRI evaluation.

1.2. Aims of the ENMC workshop

Volker Straub, UK, opened the ENMC workshop by outlining the increasing importance of MRI for the diagnosis of muscle disorders. The ENMC workshop aims are focused on 1) establishing a consensus of standardized muscle MRI acquisition protocols across centers; 2) systematically collecting and storing muscle MR images in a central repository to build international cohorts; 3) discussing scientific rigorous evaluation of muscle MRIs. Volker Straub highlighted the evolution of current diagnostic MRI protocols (lower limbs vs. whole body) and sequences (T1, STIR) [28] as well as the increasing clinical use of quantitative MRI techniques. He discussed the importance to establish and validate a secure imaging platform to share medical images between experts, and the benefits to patients by coordinating image sharing with the European reference network (ERN) EURO-NMD. He discussed that this workshop furthers the initiatives initially established by the MYO-MRI COST Action BM1304 working group 1, focused on “Improving diagnosis and understanding of muscle pathology” (https://myo-mri.eu/working-group/improve-diagnosis-understanding/).

1.3. Protocol optimization for diagnostic MRI

Anna Pichiecchio, Italy, outlined the optimization of muscle MRI protocols including defining current techniques and sequences to improve diagnostics for rare NMD [29]. Although recent studies in NMD have frequently made use of whole body imaging, many centers do not have appropriate body array MRI coils and still perform regional imaging, mainly of the pelvic girdle and lower limb muscles. For regional imaging, patient positioning is extremely important. In the lower limb assessment, correct positioning requires having feet first into the scanner with imaging up to the lumbar area, patella facing upward, lower limbs in comfortable position (feet together) on the scanner bed, sandbags and thermoplastic bands used to stabilize limbs to minimize motion. In the upper extremities, the patient is positioned head first in the scanner for the study of the cranial and scapular districts. MRI protocols vary by anatomical regions: in case of the lower limb (both legs simultaneously) and pelvic girdle muscles, axial slices are usually performed from the pelvic girdle down to the malleolus or lower if required. The axial plane has to be oriented perpendicular to the long axis of the femur and tibia respectively. When feasible, for example in children, the exam can be performed in a single field of view for thigh and calves. For the shoulder girdle, axial and coronal slices are obtained from the upper humerus to cranial muscles. Arm and forearm muscles are analyzed through axial slices from the head of the humerus down to the elbow and to the wrist/hands, for instance for distal myopathies.

Diagnostic muscle MRI protocols for patients with NMD have been reviewed previously [28,30]. Intramuscular adipose tissue assessed by T1 sequences demonstrates high signal

intensity due to the short T1 relaxation time. Several semi-quantitative scales have been published to assess the extent of adipose substitution [31,32], with the Mercuri scale being one of the most utilized [33].These semi-quantitative scales evaluate either single slices or, more recently, have been used to score the full-length of the muscle with good intra- and inter-observer agreement. However, these scores rely on subjective visual assessment and are more complex to score for intermediate grades and when the muscle is not uniformly affected. Intramuscular edema is instead characterized by means of inversion recovery (IR) T2 weighted images, where the fat signal is removed. These images are particularly useful in acute inflammatory myopathies. The challenges in STIR imaging interpretation include the lack of absolute values or standardization, in addition to the variability in fluid sensitivity of the images that differ between vendors. In addition, prolongation of T2 times have been documented in healthy muscle after exercise, acute denervation or active stages of disease progression (i.e. DMD), which may affect MR interpretation. Semi-quantitative scores for edema also exist [34-36].

1.4. Defining current diagnostic protocols

Susanna Quijana-Roy, France, outlined technical challenges with current diagnostic MRI protocols, particularly for pediatric patients. First, the pediatric population may need sedation/anesthesia in younger children and often have a time limit in the scanner of ~30–40 min. Scanning young children (shorter than 130 cm) with a 1.5 T machine using the body coil to obtain classic T1 and T2 STIR images may not lead to high quality images. Imaging quality can be improved by using a spinal-array coil instead of the standard Q-body coil [37]. Also, in difficult cases or sedated patients, prioritization of the most informative sequences (starting by T1 rather than STIR sequences in hereditary myopathies) is important if the MRI protocol needs to be terminated early due to patient agitation. Infants and young children may be scanned without sedation in adapted settings, after meals and during periods of the day when they are more prone to nap. To minimize required sedation, children can be scanned together with a parent. Additional time is required for positioning patients with severe contractures and imaging is better performed perpendicular to the long axis of extremities with contractures or with patient obesity as the arms are out of the field of view. Metallic implants (hips, spinal) can lead to imaging artifacts although most newer implant materials do not cause broad distance artifacts, allowing relatively close muscles to be visualized. Careful consideration should be taken in case of high magnetic field machines (3 Tesla or more) due to the possibility to increase the heating of tissues close to metallic implants.

Susanna Quijano-Roy also discussed the diagnostic utility of Whole Body MRI (WBMRI) and highlighted the selective pattern of muscle involvement in the genetically heterogeneous pediatric rigid spine syndrome [28,38,39]. She provided several examples of *RYR1*-core myopathy with selective signal abnormality of the soleus [40]; *SEPN1*-myopathy with absence or hypotrophy of the semimembranous muscles [41,42]; *GAA*-myopathy/Pompe disease with subscapularis and tongue muscle involvement [43,44]; ambulant *LAMA*2/merosin deficient limb girdle muscular dystrophy (LGMD R23) with brain white matter changes, subscapularis and adductor magnus muscle involvement and pseudo *COL6*-sign with tigroid texture in the lower leg [45] (Quijano-Roy et al., submitted 2020); *MYH7*-myopathy with affected axial and paravertebral muscles as well as involvement of the sartorius, tibialis anterior and soleus muscles, that may show an inverted *COL6* sign [46].

Robert Carlier, France, outlined that MRI techniques have becoming an increasingly important tool for the initial diagnosis of muscular disorders as well as for assessing progression and treatment efficacy. He highlighted the advantages of quantitative Dixon techniques, where the fat images obtained with the Dixon method are comparable to T1 images, giving similar qualitative information and simultaneously allowing to measure intramuscular fat content [37]. The 3-point Dixon technique can be used to quantify fat, total and contractile cross-sectional areas and can provide a more accurate estimate of muscle bulk that may better correlate with the patient's functional status, than simply examining cross-sectional muscle area. Although Dixon techniques require specialized software and longer analysis than traditional T1 images, quantitative Dixon results provide improved reliability between centers, which is necessary for clinical trials [47]. Typically, 3-point Dixon utilizes three acquisitions where the water and fat contributions are alternating between in phase and out of phase. In a whole-body acquisition, a network of coils including head, surface as well as coils inserted into the MRI table, permits the assessment of the entire body from head to toes in 6 consecutive stacks without requiring repositioning of the patient. The duration of this examination is 35 min for ~300, 5 mm thick images with excellent spatial resolution and signal to noise ratios [37]. In addition, radiologists can detect both brain and muscle abnormalities with this technique.

Robert Carlier also discussed combining T1 images and STIR sequences in a single IDEAL (Iterative Decomposition of water and fat with Echo Asymmetry and Least-squares) T2 sequence to decrease acquisition time, since all the image evaluation is performed on the same image slice.

1.5. Protocol optimization for quantitative muscle MRI

Pierre Carlier, France, presented on "Defining Quantitative MRI protocols to acquire WBMRI images." Traditionally, muscle diagnostic imaging is performed with different sequences and in a different environment than the longitudinal monitoring of disease progression by MRI in a clinical trial. Pierre Carlier emphasized that for disease monitoring, quantitative measurements are mandatory for a precise assessment of the fatty degenerative changes with water-fat separation (Dixon) sequences or of "disease activity" with multi TE spin echo derived water T2 maps. It has

already been suggested that both qualitative and quantitative assessment of muscle fatty replacement might be realized with one single acquisition, a whole-body Dixon scan [48]. Taking into account the wider availability of this sequence on modern scanners, and also the introduction of faster acquisition schemes, a consensus was reached within the group that whole-body Dixon imaging should be recommended as a first line imaging protocol for both diagnostic and longitudinal studies, where available. Switching to this imaging protocol will preserve the image quality and will offer the possibility to precisely evaluate the disease progression with repeat imaging.

Attention was also drawn to sequences that simultaneously evaluate both fatty degenerative changes and disease activity. Dixon sequences based on a spin echo at relatively long TE generate T2 fat fraction maps and qualitative T2 water maps. Whole-body examinations are possible with excellent anatomical delineation. Multi TE spin echo images can be decomposed into proton-density fat-fraction maps and quantitative water T2 maps. Whole-body implementation is difficult, in part because of high radiofrequency (RF) power deposition. When quantitative imaging is envisioned as a biomarker of disease progression, the question is frequently asked: which muscle is the best indicator and which muscle region should be preferentially selected? While it seems contra-intuitive, data were shown indicating that a global analysis simply discarding the subcutaneous fat had a higher or an equivalent discriminant power in a majority of diseases.

Jasper Morrow, UK, continued by providing a detailed description of the optimal analysis of quantitative MR images [49]. Data extraction from quantitative muscle MRI should be optimized to maximize outcome measure responsiveness and reduce the potential for bias. Muscle region of interest definition should be performed by a skilled and trained observer with assessment of test-retest reliability. Analysis should take place at the end of the study, with all scans from a single subject being analyzed in parallel, with the observer blinded to clinical details and scan order. Due to inhomogeneity within muscles, it is crucial to ensure the same anatomical level is analyzed on repeated assessments [50,51]. Robust quality control processes are necessary at every stage of image acquisition and analysis [52].

1.6. Building cohort studies for imaging

Giorgio Tasca, Italy, presented the recent experience of building patient cohorts for imaging, with particular regard to two large single center projects on facioscapulohumeral muscular dystrophy (FSHD) type 1 [53,54] and two multicenter projects dealing with sarcoglycanopathies [55] and FSHD type 2 [56], developed in the context of the MYO-MRI COST Action (BM1304). He underlined the importance of studying sufficiently large cohorts to understand and characterize the full variability of phenotypes and severity in rare disorders. Building international initiatives/consortia is invaluable to pursue this aim, an added importance being the exchange of ideas among experts. Giorgio Tasca highlighted that MRI is an important tool to assess asymptomatic carriers for FSHD. Even cross-sectional qualitative studies, if including a sufficient number of subjects, can contribute to improve the understanding of disease progression over time. Difficulties are mainly related to the inhomogeneity in acquisition of imaging and clinicogenetic data in retrospective studies. Therefore, standardizing imaging protocols, harmonizing data collection among the different centers, as well as having homogenous scoring systems for qualitative MRI remain critical to improving future MRI acquisition and interpretation.

John Vissing, Denmark, presented completed, ongoing and planned natural history studies for LGMD R9, FKRP-related [47,57]. A prospective, 1-year study in 32 patients with LGMD R9 showed a significant drop in Forced Vital Capacity (FVC), but no change in strength, 6 min walk test (6 MWT), timed-up-and-go test, 10 m walk test, stair descent and ascent velocity or chair rise time. By contrast, quantitative MRI showed highly significant increases in fat fraction in 9 of 14 assessed muscles [57]. A 6-year follow-up in the same patient group showed further progression in quantitative MRI findings and FVC, in addition to all of the functional measures mentioned above [58]. The findings show that quantitative MRI is a strong surrogate biomarker for clinical change in LGMD R9. Muscle contractility should also be considered in natural history studies, as contractility may change in a non-linear fashion with increasing fat fractions [59]. John Vissing also reported on an ongoing natural history study at his institution on axial muscle involvement in LGMD R9, showing that the psoas major and the lumbar paraspinal muscles are affected the most in LGMD R9 compared to FSHD, Bethlem myopathy and 4 other LGMDs. Lastly, he reported on two natural history studies performed for trial readiness, planned to start in 2020. Each study will include 60 patients. One study will be sponsored by Genethon at three sites and the other by ML Bio Solutions at over 10 sites.

1.7. Linking muscle MRI to clinical data

Jordi Díaz-Manera, UK/Spain, provided an update on the correlation between muscle function and MRI results. He reviewed the studies in which fat replacement has been analyzed using semi-quantitative scores, such as the Mercuri score. In general, he showed that there are statistically significant correlations with a high correlation coefficient (> 0.6 in most cases) between muscle fat fraction and the results of muscle function tests such as the 6 MWT, timed tests, muscle strength or also daily activity scales in Pompe disease, dysferlinopathies and also in oculopharyngeal muscular dystrophy [60-62]. Many more studies have been conducted using a quantitative approach, such as Dixon, in several other muscular dystrophies that confirm these earlier data [52]. He then described studies that have attempted to analyze whether the increase in the fat fraction detected by MRI in longitudinal studies correlates with a decrease in muscle function. The data presented showed that most of these studies have been performed in a short period of time,

usually less than a year, and in all of them the increase in the fat fraction was not associated with a decrease in the results of muscle function tests. However, he also stated that there are not enough data from longer longitudinal follow-up studies in which muscle function and muscle MRI were performed annually. There is only one follow-up study in LGMD R9 that showed that after 6 years, changes in muscle MRI are associated with worsening of the muscle function test [22,23]. Therefore, he concluded that muscle fat fraction correlates with muscle function, but more data are needed in follow-up studies to know whether increased fat fraction precedes decreased muscle function in genetic myopathies.

Jasper Morrow expanded on the correlation between MRI and clinical data. As muscle MRI provides much greater anatomical specificity than clinical assessments, therefore a perfect correlation between MRI and clinical data is not expected, or indeed desirable [63]. However, demonstrating correlation is vital to provide criterion validity to quantitative muscle MRI as a surrogate outcome measure in NMD. Quantitative MRI, in particular muscle-fat quantification, demonstrates good convergent validity (correlation between T1 and quantitative values) and has high inherent face validity. There are very strong cross-sectional correlations between MRI measures of remaining functional muscle tissue and measured isokinetic muscle strength in many diseases, including Charcot-Marie-Tooth disease and inclusion body myositis [16,49]. Overall muscle MRI severity measures, such as mean muscle fat fraction, showed good correlations with clinical measures of overall disease severity and function [31]. Recently demonstrated in patients with Duchenne Muscular Dystrophy [22,23], the next step is to show that short term changes in muscle MRI predicts long term change in patient function throughout many genetic myopathies, so that MRI can be used more widely as a bridging biomarker in clinical trials.

1.8. Correlating imaging with clinical and genomic data

Bjarne Udd, Finland, discussed correlating multiple imaging parameters with genomic data, using the examples of tititopathies. He reported on the more than 10 described distinct phenotypes of titinopathies and their genotype-phenotype correlations compared to the outcome on muscle MR-imaging. The first identified autosomal dominant late onset distal titinopathy with mutations in the last exon caused highly selective fatty replacement of the anterior lower leg muscles, variably hamstrings and gluteus minimus muscles. In homozygosity or combined with a truncating variant in the second allele (*TTN*tv), the outcome is childhood onset LGMD R10 titin-related, with fatty replacement of most lower limb muscles after age 50 [64]. In combination with recessive *TTN* A-band missense mutations, the outcome is very different with adult onset proximal lower limb weakness and more severe fatty degeneration in the quadriceps and soleus muscles. Biallelic recessive frameshift or nonsense *TTN* mutations, with at least one located in one of the two last exons, caused early or juvenile onset recessive distal myopathy with more prominent replacement of the soleus together with anterior lower leg muscles [65]. *TTN* missense mutations in exon 344 cause a totally different disease, hereditary myopathy with early respiratory failure (HMERF), and selective fatty changes in the obturatorius, semitendinosus, tibialis posterior and anterior lower leg muscles [66]. The variability of clinical and MRI phenotypes in congenital titinopathies with biallelic *TTN*tv mutations is very large, from arthrogryposis with fetal lethality, congenital amyoplasia to milder phenotypes either with or without cardiomyopathy [67]. The MRI features are correspondingly variable and complex with some diagnostic clues such as fatty degeneration of the semitendinosus muscle and adductor magnus hypertrophy.

Carsten Bönnemann, USA, presented an iterative approach to assigning plausibility to *TTN* variants in early onset recessive titinopathy. This approach used imaging together with genotype, phenotype, and histology. Analyzing patients with biallelic loss of function mutations in *TTN*, muscle MRI as well as ultrasound revealed great heterogeneity, with involvement of the proximal semitendinosus muscle as the most consistent finding across patients. This finding then helped to assign tentative *TTN* pathogenicity in patients with one predicted loss of function allele and one variant of uncertain significance (i.e., rare missense mutations) on the other allele, provided the phenotype and (if available) histology were also compatible. This approach did not require for the imaging findings to be diagnostic, but rather to be compatible with a selective pattern of pathology.

David Gómez Andrés, Spain, discussed statistical and machine learning techniques to improve pattern recognition and representation. For instance, he showed the basics of heatmaps, which are graphical representations categorizing the degree of individual muscle involvement. After the initial proposal of heatmaps for pattern representation in NMD imaging [41], they have rapidly gained popularity and acceptance [68]. Heatmaps can be easily built and provide extensive information in a comprehensive way. They can be used to delineate which muscles are part of the diagnostic ‘positive fingerprint’ (those muscles which are usually affected and that can help to guide diagnosis or support compatibility of a suspected diagnosis), or ‘negative fingerprint’ (those muscles which are never affected and that help to exclude a diagnosis) [42]. Heatmaps can incorporate semiquantitative scoring but also quantitative data [69]. Heatmaps providing graphical representation of imaging data are important for meta-analytical purposes and comparison of future results in the same or different disorders. Apart from data representation, other data mining and machine learning approaches have been incorporated to the analysis of imaging data, mainly random forests [68].

1.9. Assembling rare disease cohort MR images from the international community

Roberto Fernandez-Torron, Spain, reviewed publicly available MR images of genetic myopathies and other

NMD. In addition to the growing number of scientific papers reporting muscle MRI patterns for single diseases, there are several reviews summarizing the most common patterns of specific disorders [11,26,70]. There are also some references covering a large number of NMDs, including a detailed textbook with MRI NMD descriptions and diagrams, published by Watjjes M and Fisher D (Neuromuscular Imaging Springer Verlag New York, 2013, ISBN 978-1-4614-6551-5) and a website (https://neuromuscular.wustl.edu/pathol/diagrams/musclemri.htm). However, the most common educational resources used by clinical radiologists, such as radiopaedia (https://radiopaedia.org), lack MRI examples of muscle pathology and NMD are generally underrepresented. It was agreed by workshop participants that a comprehensive NMD MRI atlas is needed and that participants should provide examples of typical NMD MRI images to the most accessed websites by radiologists to increase awareness and as an educational tool.

Jodi Warman-Chardon, Canada, presented the MYO-Share imaging platform. MYO-Share is a secure, online imaging portal that can be used to view and compare anonymized muscle MRI scans from patients with NMD from across the world. MYO-Share was developed based on recommendations by the MYO-MRI COST Action (BM1304) consortium (https://myo-mri.eu/), which brought together top international specialists (NMD neurologists, radiologists, scientists) working towards the harmonization of diagnostic muscle imaging standards, and establish international guidelines for imaging patients with NMD. Similar to other rare disease online imaging inventories (like brain imaging atlases), MYO-Share has been developed to assemble diagnostic muscle MRI scans that are currently scattered across NMD centers internationally, to develop an inventory of images from a broad spectrum of NMD in a secure online database. MYO-Share is projected to contain thousands of anonymized MRI scans of patients with genetically confirmed myopathies to help the correct diagnosis and provide an academic resource to share characteristic images with NMD experts across the world.

Francina Munell, Spain, discussed linking images with the EURO-NMD and other platforms for the collection of clinical and phenotypic data. There is a growing list of public databases collecting phenotypic data using Human Phenotype Ontology (HPO) terms [71], but these platforms do not include imaging data. It is clear that MRI can facilitate diagnosis and follow up studies (natural history, clinical trials). There is a consensus about the need to improve HPO for NMD by developing specific terms for all NMD, including terms for histology, physiology, and possibly for imaging. The integration of data imaging in these platforms would mean a step forward in the field. On the other hand, there are still important questions to address regarding the amount of data to be included in imaging repositories (minimum phenotypic data, dynamic phenotyping or deep phenotyping) and if the data should be common or specific for each disease group. Development of systems that allow exporting data from existing local databases would facilitate this work.

1.10. Recommendations and workshop deliverables

1.10.1. Recommended protocols

The workshop participants agreed that the "standard" protocols for MRI are continuing to evolve. Currently, Dixon techniques provide better quantification of muscle content of water and fat per pixel and, when possible, should be chosen to quantitatively assess fatty tissue replacement in muscle [30]. T1 images should be used to assess fatty replacement if quantitative techniques are not available. T1 are semi-quantitatively assessed and several scores have been proposed [32,36,72]; modified Mercuri scores in either 4 or 5 points being most commonly used in the consortium. To assess early inflammation and edema, T2 images with fat saturation and STIR images complement Dixon/T1 imaging. Participants also discussed current limitations of the diagnostic techniques. For example, muscle hypotrophy may not always be obvious and measures of muscle volume can be challenging. Interpreting mild increases in STIR signal can also be difficult. Even in healthy controls, mild increases in STIR signals in the calf muscles can occur with minimal activity. The use of contrast agents in muscle imaging is not recommended for routine diagnostics of genetic NMD. Centers need to incorporate evolving imaging techniques to reduce acquisition time and to simultaneously acquire fat and water images comparable to STIR sequences with better resolution, such as Dixon sequences based on a spin echo at relatively long TE.

Workshop participants agreed that WBMRI should be applied when possible. If unavailable, dedicated protocols to study body regions of interest in addition to the standard lower limb assessment should be used. An additional benefit of WBMRI is the detection of possible abnormalities in abdominal and thoracic organs. Finally, for patients with neurocognitive features, brain MRI should be added to whole-body imaging to identify malformations, leukodystrophy or ocular involvement, which are more commonly associated with congenital muscular dystrophies [73,74]. The workshop participants agreed that imaging larger disease cohorts is important to better characterize diagnostic imaging patterns. Even in diseases where there is no diagnostic indication for muscle imaging, as e.g. in Duchenne muscular dystrophy, the application of muscle MRI is still extremely important for natural history studies or as an outcome measure in clinical trials.

1.10.2. MYO-Share and expand muscle MRI cohorts

Currently, there are several published cohorts shared by many of the consortium members [55,58,61,62,75], however, no single platform has been used to share images internationally. The ENMC workshop participants discussed expanding international collaborations to build more comprehensive cohorts of specific NMD, facilitated by an imaging-sharing platform. The MYO-Share platform has been recently developed as a proof of concept to rapidly increase the sharing and contribution of muscle MRI scans. This infrastructure would facilitate the prospective cohort

development for multiple different countries and would allow the collection of images from age-matched healthy controls. To assess the feasibility of the MYO-Share platform, the ENMC workshop participants decided to initially start with a few selected diseases (*ANO5*-related LGMD R12 being the first) with selected clinical information collected. It was also discussed to share a common MYO-Share ethics proposal to be modified for each research center to assist with the ethics submission requirements. Additional challenges remain in ensuring the strictest privacy and consent protocols for uploading, sharing and storage of images, given the variation in privacy and ethics requirements in each country. In order for the sustainability of MYO-Share, future meetings will be held to develop a governance committee and a scientific review committee to integrate novel imaging techniques as well as to address sustainability including linking with patient organizations and industry. Pierre Carlier also discussed the opportunity of integrating MYO-Share with EURO-NMD. Selected anonymized images could be archived in MYO-Share and be integrated as representative teaching cases. Further discussions will be held on how to link the ERN platform to MYO-Share.

1.10.3. NMD imaging atlas

Unfortunately, many of the muscle MR images that are freely available online do not accurately reflect the characteristic diagnostic patterns that are described in the full literature. In order to improve the dissemination of characteristic NMD MRI patterns, an updated imaging atlas with selected images needs to be created. The workshop participants agreed on the need for a well-designed NMD atlas curated by NMD imaging specialists. In the atlas, the key muscles at different anatomic levels should be highlighted for each disease subtype. The group discussed that the atlas will be publicly available and housed within MYO-Share. The atlas should include as a minimal requirement the age of the patient, disease stage, focus on important muscles affected and spared late in the disease. The ENMC workshop participants agreed that a scientific committee should supervise key selected images presented to ensure accuracy of the images provided in the atlas. This NMD imaging atlas would also include images of normal muscle anatomy and age matched controls.

1.10.4. Future directions in muscle MRI

With hundreds of genetic myopathy subtypes, machine learning (ML) may facilitate the diagnostic assessment. In a recent proof-of-concept study, ML has been demonstrated as highly effective to identify patterns of pathology and specific discriminatory muscles of lower limb MRI involvement in muscular dystrophies. The authors also made the algorithm available to classify future patients [76]. Interestingly, the most discriminatory muscles lay within the pelvic girdle and frequently were not included in the classic MRI patterns of assessment. Moving this promising method forward will require increasing the number of images assessed, necessitating larger international collaborations with shared imaging databases [77]. In order to decrease the burden of the visual scoring of each muscle, automated muscle segmentation is needed. Systematic comparison between different diseases, at various ages and a range of severity of these conditions will be required to ascertain the diagnostic value of MRI in this context [78]. ML may also provide the technology to link big genomic data to those provided by muscle imaging.

2. Conclusion and workshop deliverables

This workshop assembled international experts with the goal of harmonizing MRI protocols and to strategically plan the building of future cohorts with genetic muscle diseases. The workshop also highlighted the importance of developing a systematic, consensus-based imaging approach that will be coordinated by the MYO-MRI consortium in close collaboration with EURO-NMD. This coordinated approach will enhance data collection and collaboration that complements the research being carried out in the individual centers. The workshop reviewed optimizing online imaging portals, such as the MYO-MRI MYO-Share imaging platform and the development of an online imaging atlas.

In conclusion, muscle MRI is now becoming an integral part of the diagnostic algorithm for most myopathies, to distinguish between phenotypically similar disorders, guide interpretation of genetic testing, and as a biomarker of disease progression or response to therapies. Future meetings are planned to facilitate building larger disease cohorts, integrating a common imaging platform, and applying emerging technologies such as ML to assess whether this will help differentiate disorders where MRI is not yet considered useful or diagnostic.

3. Workshop participants

-Carsten G. Bönnemann, Bethesda, USA.
-Pierre G. Carlier, Paris, France.
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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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