Abstracts from Theme SW and CW

Scientific Work in Progress

Clinical Work in Progress and Care Practice

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THEME SW: SCIENTIFIC WORK IN PROGRESS

SW1 A NEFH GENE MUTATION MANIFESTING ALS-FTD COMPLEX: FIRST KOREAN FAMILIAL ALS-FTD

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Keywords: amyotrophic lateral sclerosis, frontotemporal dementia, neurofilament

Introduction: The etiology of the majority of amyotrophic lateral sclerosis (ALS) remains unknown. Several findings from some laboratories suggest a role for neurofilaments in the development of motor neuron disorders and indels of the heavy neurofilament subunit (NEFH) tail have been associated with ALS. Recently, we have identified deletion mutation of the NEFH gene in patients with familial ALS and frontotemporal dementia (FTD) complex.

Case study: A 53-year-old male, who had familial history of muscular weakness and dementia, presented cognitive decline and slowly motor weakness approximately fifteen months ago, and was diagnosed with familial ALS-FTD complex. Genetic analysis with ALS-related known mutation of the Human Gene Mutation Database (HGMD) in the patient revealed a mutation (p.Ser752_Lys757del) in NEFH gene.

Discussion: Although previous findings demonstrated that NEFH gene mutation showed predominantly sporadic ALS rather than FTD, but the patient and family members in this study presented ALS-FTD complex. These findings suggested that the NEFH gene mutation would be a plausible mutation in ALS-FTD complex in Korea.

References:

1. Boylan KB, Glass JD, Crook JE, et al. Phosphorylated neurofilament heavy subunit (pNF-H) in peripheral blood and CSF as a potential prognostic biomarker in amyotrophic lateral sclerosis. Journal of neurology, neurosurgery, and Psychiatry 2013;84:467-472.

2.Al-Chalabi A, Andersen PM, Nilsson P, et al. Deletions of the heavy neurofilament subunit tail in amyotrophic lateral sclerosis. Human Molecular Genetics 1999;8:157-164.

3.Figlewicz DA, Krizus A, Martinoli MG, et al. Variants of the heavy neurofilament subunit are associated with the development of amyotrophic lateral sclerosis. Human molecular genetics 1994;3:1757-1761.

4.Skvortsova V, Shadrina M, Slominsky P, et al. Analysis of heavy neurofilament subunit gene polymorphism in Russian patients with sporadic motor neuron disease (MND). European journal of human genetics : EJHG 2004;12:241-244.

Acknowledgments: This study was supported by a grant from the Korea Healthcare Technology R and D Project, Ministry for Health, Welfare and Family Affairs, Republic of Korea (A120182).

SW2 AMYOTROPHIC LATERAL SCLEROSIS INCIDENCE AND CLUSTER IDENTIFICATION IN NEW BRUNSWICK, CANADA, OVER A 10-YEAR PERIOD

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Keywords: New Brunswick, Cluster, Incidence

Background: To date no studies on Amyotrophic Lateral Sclerosis (ALS) in the Canadian province of New Brunswick (NB) have been conducted. New Brunswick (population 735,000) has European and First Nations ancestry, with Acadian, Irish and British predominance, with many communities having little migration over the centuries. Epidemiological studies are crucial to study rare diseases; they inform basic science studies and provide information on sociodemographic characteristics, disease phenotype, geographical variations, risk factors and protective factors (1).

Objective: The purpose of this study was to perform a 10 year retrospective epidemiologic study of ALS in NB, identifying unique patterns or clusters that could contribute to our knowledge about the disease.

Methods: A database of patients diagnosed with ALS between January 2003 and December 2013, was created from the health records of the Stan Cassidy Centre for Rehabilitation (SCCR). The SCCR has an established provincial ALS clinic, following approximately 60 patients at any time. Data extracted included: date of symptom onset, date of diagnosis, age and pattern of disease onset. Addresses were assigned to their Canadian 2011 Census Subdivisions and plotted in the ArcGIS© spatial analysis program to assess for potential clusters, with data adjusted for age and population density to the Canadian 2011 Census data.

Results: 187 patients diagnosed with ALS within the 10-year period were identified. Average yearly incidence was 2.50/100,000. Average age at diagnosis was 65.0. More males (56%) (Chi Square= 4.143; P-Value= 0.0418) were identified. Average time between onset of symptoms and diagnosis was 1.4 years. (n=103). Limb onset was 57% and Bulbar onset 35% of all cases. Familial ALS was characterized by 8% of the total ALS population. The cluster analysis suggested four areas of high incidence of ALS cases; Bathurst, Edmundston, St. Paul and Wilmot area. Each cluster is defined as having at least 95% confidence interval.

Discussion and conclusion: Even as a known under-representation of the New Brunswick population, this study suggests that New Brunswick has a higher incidence of ALS compared to recent incidence counts published from other Canadian provinces. The study also supports recent literature suggesting Males are more susceptible to ALS than Females. The proportion of bulbar onset cases compared to the other sites of onset make up over one third of all onset cases. This is significantly higher than the globally accepted value of a quarter. Reasons for this discrepancy should be studied in future research. In this ongoing study, the database will be expanded to by integrating datasets from the provincial Medicare, Vital Statistics, hospital discharge and the ALS Society of New Brunswick.

References:

1. Factor-Litvak *et al.* Amyotrophic Lateral Scler.Frontotemporal Degener. 2013; 14: 33-43.

SW3 ENTEROVIRAL INFECTION CAUSES CYTOPLASMIC AGGREGATION AND CLEAVAGE OF TAR DNA BINDING PROTEIN-43

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Keywords: TDP-43, enterovirus

Coxsackievirus B3 (CVB3) is a non-enveloped, positive single-stranded RNA enterovirus that infects the brain, pancreas, and heart. The etiology of amyotrophic lateral sclerosis (ALS) has been speculated to be partially due to enteroviruses, including Poliovirus and CVB3, however the current literature remains controversial as to whether enteroviruses are present in a subset of ALS patients. Moreover, the pathophysiology of enterovirus infection has never been investigated to possibly induce ALS-like pathologies. Growing literature suggests that the molecular pathologies between enterovirus-induced diseases and neurodegenerative diseases share a similar cellular pathophysiology. In ALS, insoluble-cytoplasmic TAR DNA binding protein-43 (TDP-43)-aggregates are a common biomarker contributing directly to pathological disease progression. TDP-43 is an RNA-binding protein and plays an essential role in regulating RNA metabolism at multiple levels. The importance of insoluble protein aggregates in enteroviral pathogenesis has also been widely recognized, resembling that of neurodegenerative diseases. However, the significance of TDP-43 in CVB3-infection remains unstudied.

In this study, we investigated the pathological similarities between CVB3-infection and ALS, with a focus on TDP-43. Our studies demonstrated that CVB3 infection leads to redistribution of nuclear TDP-43 into cytoplasmic aggregates in a viral protease 2A dependent manner. We also found that viral protease 3C, actively cleaves TDP-43 at amino acid 327 into two respective cleavage fragments. As a consequence of cleavage, the respective N-terminal cleavage fragment inhibits native TDP43 activity in the nucleus as a transcriptional regulator. We conclude that TDP-43 pathologies in CVB3-induced pathologies may be similar to those found in ALS.

SW4 ACCRUED SOMATIC MUTATIONS TRIGGER ONSET OF AMYOTROPHIC LATERAL SCLEROSIS: FROM HYPOTHESIS TO MAINSTREAM THINKING (2005-2015)

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Keywords: disease onset, Somatic Mutations, nucleic acid changes

In 2005 I brought together clinical and epidemiological evidence to support the hypothesis that acquired nucleic acid changes may trigger sporadic ALS (1). The article arose from the understanding (a) of ALS as a 2-3 level disease of the motor neuron system, and (b) that the mechanisms triggering disease onset needed to be considered separately from those facilitating its spread within the motor neuron system and from those causing the demise of the motor neurons. Twelve principal building blocks underlay this hypothesis: 1. Understanding ALS as a Neural Network Disease; 2. Separation of ALS Onset, Spread, and Motor Neuronal Demise; 3. Clinical Clue - Pattern of Spread; 4. Clinical Clue - Individualized rate of progression; 5. Clinical Clue - Association with MGUS; 6. Familial ALS Clue - Most fALS is AD; 7. Familial ALS Clue - Incomplete penetrance; 8. Familial ALS Clue - Phenotypic heterogeneity; 9. Familial ALS Clue - Phenotypic differences between AD ALS (rapidly progressive) and AR ALS (indolent); 10. Clues from western Pacific ALS; 11. Clue from sporadic disease - incidence increases with age; 12. Clue from sporadic disease - smoking as a risk factor.

Most of the conceptual foundations for this hypothesis have been strengthened during the past ten years. The ALS journal was renamed "ALS and FTD," in recognition of the continuity of the motor system network, to include the part of the brain where motor activity is planned. The focal onset, with simultaneous initial maximal upper and lower motor involvement in the region of onset, and patterns of spread, have been characterized further. Clues from the epidemiology of sporadic ALS have been affirmed by additional data and by quantitative analysis: the increase in disease incidence with age suggests accrual of time-dependent changes, and smoking, a known carcinogen, has been affirmed as an established risk factor. The implications of the phenotypic heterogeneity and of the incomplete penetrance of familial ALS have been realized.

As a result, the understanding that accrued somatic mutations trigger onset of ALS is ready to become mainstream thinking.

Reference:

1. Armon C. Muscle and Nerve 2005:32:373-377.

SW5 CORTICOSPINAL MOTOR NEURON DEVELOPMENT CONTROL GENES AS CANDIDATES FOR HUMAN ALS SUSCEPTIBILITY

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Keywords: SOX5, Burden testing, Rare Variants

Introduction: A key problem in genetics is the interpretation of genetic variation and whether it is relevant to disease. This is particularly an issue for ALS in which the genetic architecture is complex. Current evidence suggests some ALS is caused by a few common variants, such as the hexanucleotide expansion in C9orf72, but most is caused by many rare variants. A candidate gene and convergent approach is required to identify rare variation confidently. We therefore studied motor neuron developmental genes, and combined gene-level, rare variant, haplotypic and gene expression analyses to identify disease-causing genes.

Method: We used samples from three post-mortem datasets: genotype data (n = 5619 and n = 9230) imputed for rare variants, gene expression (6 ALS cases; 3 controls), and DNA-sequence data (50 ALS cases and 88 controls). We selected 28 candidate genes based on corticospinal motor neurons (CSMN)

regulation, genes involved in CSMN axonal extension and genes associated with cellular processes associated with ALS. We ran multiple gene-level burden tests (eg SKAT), haplotype association analyses, logistic and linear regressions of rare variants predicting disease status and other ALS phenotypes, epistasis analyses between variants, and differential gene expression analyses. Statistical p-values were permutated and corrected by 28 tests. Unadjusted p-values are reported.

Results: Logistic regression showed statistically significant association across three genotype datasets for SRY (sex determining region Y)-box 5 (SOX5) with ALS. (rs1479443 OR: 0.81 and p-value: 4.29x10-6), which contributed to a haplotype statistically associated with ALS cases (highest p-value 6.10x10-6). SOX5 showed significant differential expression when comparing ALS affected and control spinal cord tissue-derived total RNA (log2 Fold-Change = 0.09, p-value = 5.5x10-5), most significant in thoracic samples (log2 FC = 0.13, p-value = 6.6x10-4). Using DNA-seq data a gene-level burden test on ALS also showed association (p-value = 8.94x10-12). The cysteine rich transmembrane BMP regulator 1 (CRIM1) gene revealed gene-level association with ALS using combined-variants SKAT test (p-value = 5.67x10-4). This was confirmed, showing both significant association across all datasets for individual variants (rs6544006, OR: 0.77, p-value = 3.77x10-5), and a statistically significant ALS-associated haplotype (p-value = 2.84x10-6). Variants in the cadherin 13 (CDH13) gene showed significant association across two genotype datasets (chr16:82733062, OR: 0.66, p-value = 1.94x10-5), which formed a statistically significant ALS-associate haplotype (p-value = 0.004).

Discussion: Analysing genes that regulate motor neuron development has revealed disease-causing variation across multiple genetic modalities in SOX5 and CRIM1, and supports the use of a candidate gene approach for gene discovery in ALS.

Acknowledgements: This work is directly funded by the ALS Association (Investigator-initiated grant 14-LGCA-182 to AAC and JDM) and the Motor Neurone Disease Association of Great Britain and Northern Ireland (top-up grant for additional experiments), and by a separate ALS Association Investigator-initiated grant Q619D6 to JDM. This work was also supported by a Medical Research Council PhD studentship awarded through the MRC Centre for Neurodegeneration Research and by the European Community's Health Seventh Framework Programme (FP7/2007–2013) 259867, and a grant (075615/Z/04/z) from the Welcome Trust. This project was supported by ZonMW, under the frame of E-Rare-2, the ERA-Net for Research on Rare Diseases. This is an EU Joint Programme - Neurodegenerative Disease Research (JPND) project. The project is supported through the following funding organisations under the aegis of JPND - www.jpnd.eu: Belgium, The National Funds for Scientific Research (F.R.S. FNRS); France, Agence Nationale de la Recherche (ANR); Germany, Bundesministerium für Bildung und Forschung (BMBF); Italy, Ministero della Salute; Italy, Ministero dell'Istruzione, dell'Università e della Ricerca (MIUR); The Netherlands, The Netherlands Organisation for Health Research and Development (ZonMw); Sweden, Swedish Research Council (VR); Switzerland, Schweizerischer Nationalfonds zur Förderung der wissenschaftlichen Forschung (SNF); United Kingdom, Medical Research Council (MRC).This work was also supported secondarily by NIH grants R01NS075672, R01NS045523, and R37NS41590 to JDM.

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SW6 ONSET OF MND WITH ISOLATED UNILATERAL TEMPORALIS AND MASSETER MUSCLE WASTING: ASSOCIATION WITH ARG521CYS FUS GENE MUTATION

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Keywords: FUS, temporalis, masseter

Background: MND can present with limb, bulbar or respiratory muscle weakness. Isolated unilateral temporalis and masseter muscle wasting has not previously been described as a presenting feature. FUS gene mutations are associated with MND/frontotemporal dementia overlap syndromes, but are also well-described in patients with early

onset familial MND without cognitive involvement (1).

Objectives: To define the clinical, radiological and genetic features of a 49 year old woman who presented with unilateral temporalis and masseter muscle atrophy.

Methods: The initial presentation and clinical course of the patient and her family history are described. Magnetic resonance images of her head were acquired. Genetic testing for common familial MND mutations, nerve conduction studies, electromyography and left quadriceps muscle biopsy were carried out. The post-mortem histology report of her mother was reviewed.

Results: The patient presented with left-sided temporalis and masseter atrophy. She had difficulty opening her mouth and pain on attempted chewing. She experienced paraesthesiae in her upper limbs; sensory examination revealed reduced temperature sensation in the lower legs. Reflexes were depressed. Cognitive function was normal. Over ten months, the muscle atrophy spread to the right temporalis and masseter, she became aware of twitching in her legs and her left arm, and her left arm became weak. She lost 6kg in weight. Magnetic resonance imaging demonstrated the initial focal muscle involvement. There was no overt brain atrophy or increased signal in the corticospinal tracts. Neurophysiology at presentation was inconclusive, showing acute denervation activity in the left arm and leg and the right temporalis and biceps, and borderline sensory action potentials. Left quadriceps histology showed features of denervation. A known pathogenic Arg521Cys mutation was detected in the patient's FUS gene. SOD1, C9ORF72 and TARDBP testing was normal.

Our patient's mother had been diagnosed with MND, having presented with neck weakness followed by swallowing problems and arm weakness. Her post-mortem showed depletion of anterior horn cells in the spinal cord and depletion of motor neurons in the vagal, hypoglossal and trigeminal motor nuclei. Two other family members had histories suggestive of MND.

Discussion and conclusions: This is the first description of MND presenting with unilateral temporalis and masseter muscle atrophy. The case expands the known phenotype of FUS gene mutations. We considered a diagnosis of facial onset sensory motor neuronopathy (FOSMN) when this patient first presented although the minimal sensory signs would have been atypical. This case is further evidence of the overlap between FOSMN and MND (2,3), with the clinical features and family history in this case bringing it closer to MND.

References:

1. Vance C, Rogelj B, Hortobagyi T *et al.* Science 2009; 323:1208-1211.
2. Broad R, Leigh P. Pract Neurol 2015, 15:4 293-297.
3. Dalla Balla E, Rigamonti A, Mantero V *et al.* J Neurol Neurosurg Psychiatry 2014; 85:1009-1011.

SW7 C9ORF72 PROMOTER HYPERMETHYLATION IS LOST IN REPROGRAMMED ALS PATIENT CELLS AND RE-ACQUIRED DURING MOTOR NEURON SPECIFICATION

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Keywords: C9Orf72, hypermethylation, cytosine hydroxymethylation

Amongst several genetic mutations known to cause amyotrophic lateral sclerosis (ALS), a hexanucleotide repeat expansion in the C9orf72 gene is the most common. In approximately 30% of cases, increased cytosine methylation (5mC) of the C9orf72 promoter is observed resulting in a less severe phenotype. The developmental timing of C9orf72 promoter hypermethylation and the reason why it occurs in only a fraction of patients remains unknown. In order to model the acquisition of C9orf72 hypermethylation and examine the role of cytosine hydroxymethylation (5hmC), we generated induced pluripotent stem cells (iPSCs) from a rare ALS patient with C9orf72 promoter hypermethylation. Our data show that 5mC levels are dramatically reduced upon reprogramming and re-acquired during neuronal specification, while 5hmC levels increase following reprogramming and are highest in iPSCs and neurons. Our studies show that iPSCs are a valuable model system for examining C9orf72 heterochromatinization and reveal a potential role for cytosine demethylation.

SW8 ALS/FTD LINKED POLYDIPEPTIDES PRODUCED BY RAN TRANSLATION OF HEXANUCLEOTIDE REPEAT-CONTAINING RNAS ENCODED FROM THE C9ORF72 GENE ACCUMULATE MOST IN UNAFFECTED BRAIN REGIONS

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Keywords: neuropathology, dipeptide repeats, C9orf72

Background: In 2011, it was shown that an abnormal expanded hexanucleotide repeat (GGGGCC) in intron 1 of chromosome 9 open reading frame 72 (C9orf72) is the most common genetic abnormality in familial ALS and FTD cases (1, 2). This sense repeat expansion (GGGGCC) and its corresponding antisense repeat expansion (CCCCGG) result in the production of sense and antisense RNA foci (3). Bidirectional transcription and repeat-associated non-ATG (RAN) translation of these RNA foci can cause the production of six dipeptide repeat proteins (DPRs). Dipeptides translated from the sense strand include: poly Gly-Pro with unique C-terminal (poly GP-CT), poly Gly-Ala (poly GA), and poly Gly-Arg (poly GR). Antisense strand dipeptides include: poly Gly-Pro (poly GP), poly Ala-Pro (poly AP), and poly Pro-Arg (poly PR). All six dipeptides have been observed in CNS material of C9orf72 cases, but dipeptides originating from the sense stand are more frequent than antisense-related dipeptides (3). Dipeptide repeat protein aggregates can be seen in different parts of CNS including disease and non-disease related areas (4).

Objectives: The goal of this study is to achieve a more comprehensive evaluation of DPRs in the brain and spinal cord of C9orf72 patients. Specifically, we will determine if DPR burden is greater in disease-related regions.

Methods: With the use of immunohistochemistry and immunofluorescence, we are evaluating the presence of 5 different DPRs: GA, GP, GR, PA, and PR in various regions of human brain (motor cortex, frontal cortex, retrosplenial granular cortex, hippocampus, and cerebellum) and spinal cord (cervical, thoracic, and lumbar) of five C9 positive ALS cases, five sporadic ALS cases matched for age and site of disease onset, and five age-matched controls.

Results: Experiments and data analysis are ongoing. But we have found that DPRs aggregations are more prevalent in areas that seem not related to disease symptoms like cerebellum and hippocampus.

References:

1. Renton AE, Majounie E, Waite A, et al. A hexanucleotide repeat expansion in C9ORF72 is the cause of chromosome 9p21-linked ALS-FTD. Neuron 2011;72:257-268.
2. DeJesus-Hernandez M, Mackenzie IR, Boeve BF, et al. Expanded GGGGCC hexanucleotide repeat in noncoding region of C9ORF72 causes chromosome 9p-linked FTD and ALS. Neuron 2011;72:245-256.
3. Zu T, Liu Y, Bañez-Coronel M, et al. RAN proteins and RNA foci from antisense transcripts in C9ORF72 ALS and frontotemporal dementia. Proceedings of the National Academy of Sciences 2013;110:E4968-E4977.
4. Mackenzie IR, Frick P, Neumann M. The neuropathology associated with repeat expansions in the C9ORF72 gene. Acta neuropathologica 2014;127:347-357.

SW9 INCREASED PHOSPHORYLATED NEUROFILAMENT HEAVY (PNF-H) IN CSF AS A POTENTIAL DISEASE MARKER OF CANINE DEGENERATIVE MYELOPATHY

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Keywords: neurofilament, SOD1, biomarker

Background: Phosphorylated neurofilament heavy (pNF-H), a major structural protein of myelinated motor axons, has shown promise as a prognostic biomarker in diseases of the nervous system such as multiple sclerosis and amyotrophic lateral sclerosis (ALS) (1, 2). Canine degenerative myelopathy (DM) is an adult-onset neurodegenerative disorder resulting from mutations in *superoxide dismutase-1* (*SOD1),*

with similarities to upper motor neuron onset familial ALS (3). The clinical spectrum of DM is homogeneous within and across breeds. DM initially manifests as spastic upper motor neuron hindlimb weakness and ataxia (stage 1). Progressive weakness results in the inability to walk (stage 2) and involvement of the forelimbs (stage 3). End-stage disease culminates in flaccid paralysis of all limbs, widespread muscle atrophy and signs of brainstem dysfunction (stage 4). We aim to test the hypothesis that pNF-H will be increased in the CSF of DM-affected dogs at each disease stage relative to control dogs.

Objective: The goal of this current work is to quantify the concentration of pNF-H in the CSF of DM-affected dogs throughout disease progression and compare these finding to age-matched, neurologically normal dogs. Furthermore, we aim to compare CSF pNF-H concentration between aged, normal dogs and dogs with other chronic spinal cord disease.

Methods: Using a mouse monoclonal sandwich enzyme-linked immunosorbent assay (ELISA), we measured pNF-H concentration in CSF from aged control dogs (n=5) and DM-affected dogs (stage 1 n=4; stage 2 n=5; stage 3 n=4); data were analyzed via ANOVA with *post hoc* Holm-Sidak method. CSF pNF-H concentration was compared between aged control dogs (n=5) and dogs with other chronic spinal cord disease (n=3); data were analyzed via students t test.

Results: DM-affected dogs at each disease stage exhibit a robust increase in CSF pNF-H compared to control dogs ($p = 0.013$); no differences in CSF pNFH were observed between DM disease stages ($p = 0.704$). There was no difference between aged control dogs and dogs with other chronic spinal disease ($p = 0.741$).

Conclusions: These preliminary findings suggest that evaluation of pNF-H in CSF is potentially a sensitive and specific disease marker for DM. These data warrant further study of pNF-H in CSF and serum for diagnosis of disease and longitudinal monitoring of therapeutic efficacy in DM.

References:

1. Gresle MM, Liu Y, Dagley LF *et al* JNNP 2014; 85(11):1209-13
2. Boylan KB, Glass JD, Crook JE *et al* JNNP 2013; 84(4):467-72
3. Awano T, Johnson GS, Wade CM *et al* PNAS 2009;106(8):2794-9

Acknowledgements: AKC Canine Health Foundation (Grant #2165) and the American Boxer Charitable Foundation

SW10 INCREASED IN VIVO GLIAL ACTIVATION IN PATIENTS WITH PRIMARY LATERAL SCLEROSIS (PLS) ASSESSED WITH [11C]-PBR28 POSITRON EMISSION TOMOGRAPHY

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Keywords: PET, PBR28, PLS

Background: Multiple lines of evidence implicate the neuroimmune system in the pathology of amyotrophic lateral sclerosis (ALS). We recently demonstrated increased in vivo glial activation in the motor cortices of people with ALS by using [11C]-PBR28 positron emission tomography (PET). Glial activation correlated with disease severity and upper motor neuron burden scores (1). These findings suggest that [11C]-PBR28 may be a potential marker of upper motor neuron dysfunction in ALS. Thus, it may also represent a candidate biomarker for motor neuron disease that primarily affects the upper motor neurons such as primary lateral sclerosis (PLS). However, [11C]-PBR28 binding has not been studied in PLS so far.

Objectives: To assess [11C]-PBR28 binding in people with PLS.

Methods: Four patients with PLS and four age- and [11C]-PBR28 binding affinity- matched healthy controls completed a [11C]-PBR28 PET scan. Standardized uptake values were calculated from 60-90 min post-injection and normalized to whole brain mean.

Results: Voxelwise whole brain analysis showed increased [^{11}C]-PBR28 binding in the motor cortices in patients with PLS compared to healthy controls ($Z>2.3$, cluster corrected at $p<0.05$). The pattern of [^{11}C]-PBR28 binding in people with PLS was similar to the one observed in a previous study in ALS (1).

Conclusions: Increased in vivo glial activation is present in the motor cortices of both PLS and ALS patients. These results suggest that glial activation may be implicated in both conditions. Studies are ongoing to characterize the longitudinal changes in [11C]-PBR28 binding in both populations and their correlation with disease severity and upper motor neuron burden.

Reference:

1. Zürcher et al. Increased in vivo Glial Activation in Patients with Amyotrophic Lateral Sclerosis: assessed with [11C]-PBR28. Neuroimage Clin. 2015. Jan 19;7:409-14.

SW11 ULTRA-HIGH FIELD MAGNETIC RESONANCE SPECTROSCOPY IN ALS

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Keywords: biomarkers, magnetic resonance spectroscopy, imaging

Background: Magnetic resonance imaging has promise as a biomarker in ALS because of its ability to gather multimodal information in a non-invasive fashion. In particular, magnetic resonance spectroscopy (MRS) allows noninvasive investigation of neurochemical changes in the brain, which has potential value both as a marker of disease severity and as an opportunity to study the pathophysiology of ALS *in vivo*. Glutathione (GSH) deficits are a marker for oxidative stress, which has been implicated in motor neuron degeneration in ALS. Decreased motor cortex GSH levels in ALS have been reported at 3 T using edited MRS but remain to be confirmed with short echo MRS that will not be confounded by T_2 changes. Diffusion MRI (dMRI) can provide insight into axonal integrity in the corticospinal tract and other affected regions but has demonstrated limited sensitivity to date. Its value as a longitudinal marker of disease progression is unknown. High angular resolution diffusion imaging (HARDI) offers the potential to increase our sensitivity for structural change in cross-sectional investigations across subjects and in longitudinal investigations within subjects.

Objectives: 1) To compare GSH concentrations in motor cortex using an edited 3 T protocol with a non-edited 7 T method that would eliminate possible T_2 effects; 2) To utilize the sensitivity of MRS at 7 T to assess other neurochemical changes in ALS in both cross-sectional and longitudinal studies; 3) To apply HARDI to a cross-sectional ALS cohort to investigate its potential for improved sensitivity.

Methods: Subjects with ALS and healthy control subjects underwent edited MRS of motor cortex and dMRI at 3 T, and MRS of motor cortex and pons at 7 T. Scans were performed at study enrollment and longitudinally for up to 12 months in some subjects with ALS and in a subset of healthy control subjects. All subjects underwent a neuromuscular examination at enrollment as well as ALSFRS-R and ECAS at baseline and all longitudinal visits.

Results: Preliminary baseline data demonstrates a strong trend for lower glutamate in the pons (p=0.06) between subjects with ALS (N=6) and controls (N=5). The modest difference likely reflects a relatively high ALSFRS-R score at enrollment (39 ± 5, range 27 - 45) when compared with prior studies, indicating low disease burden at enrollment, as well as small effect size. Importantly, longitudinal data in a small ALS cohort (N=5) indicate a reduction over ~7 months in motor cortex total NAA and GSH corresponding with change in ALSFRS-R, suggesting that they may be good biomarkers of progression if baseline studies are performed early. Baseline edited 3T GSH findings are consistent with 7T GSH findings. Baseline and longitudinal change in metabolites of interest in a larger cohort as well as HARDI data will be presented.

SW12 EVALUATING BIOMARKERS IN ALS EFFICACY TRIALS

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Keywords: biomarkers, surrogate biomarker

Background: Biomarkers have gained increasing attention in recent years. Lack of effective biomarkers is recognized as a key limitation in progress to develop curative therapies in ALS. New measures are frequently promoted as biomarkers, including changes in strength, nerve conduction and

impedance, analytes in blood and CSF, structural, functional, and spectroscopic imaging of the brain, and other biophysical traits that are believed to be unique to ALS or measure ALS disease progression. Multiple studies are being conducted to evaluate and validate new biomarkers. Most applications for funding require biomarkers of target engagement and as proxies for clinical efficacy. Nevertheless, discussions of biomarkers are confused and proposals for evaluating the utility of biomarkers are often misguided.

Objective: Our aim is to provide standards for describing and evaluating biomarkers in ALS clinical research.

Methods: We attempt to advance the field in two ways. Firstly, we borrow from a large body of existing literature to identify distinct classes of biomarkers and their uses, differentiating diagnostic, prognostic, and surrogate biomarkers. Secondly, we exploit the tools of causal analysis to provide a concrete framework for understanding the value of biomarkers, particularly in the context of evaluating efficacy of new treatments, thereby defining the features of biomarkers that determine their utility as proxies for treatment efficacy.

Conclusions: We show that common metrics for evaluating biomarkers can lead to poor choices. We present study designs that would be required for validation of a biomarker used to evaluate efficacy of new therapies. We acknowledge our current limitations in identifying good biomarkers both due to the heterogeneous pathophysiology of ALS and due to the lack of existing effective therapies.

SW13 NEW INSIGHTS INTO NOVEL PROGNOSTIC BIOMARKERS OF LONGEVITY IN ALS PATIENTS

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Keywords: diagnostic biomarker, prognosis biomarker, lymphocyte

Background: Previous studies in our group analyzed expression level of seventeen genes (*Ankrd1, Calm1, Col19a1, Fbxo32, Gsr, Impa1, Mef2c, Mt2, Myf5, Myod1, Myog, Nnt, Nogo A, Pax7, Rrad, Sln* and *Snx10*) in muscle biopsies from the transgenic *SOD1G93A* mice. Our findings suggested five potential prognostic biomarkers of longevity in this animal model (*Mef2c, Gsr, Col19, Calm1 and Snx10*) (1). In spite of the fact that the search of biomarkers in ALS is being carried out in a wide variety of patient's samples, growing tendency relies on the study of new and less invasive tissues (2,3).

Objectives: In order to translate this study to human samples, our main aim is to validate the diagnostic and prognostic capacity of the selected genes in human samples, especially the blood.

Methods: cDNA serial samples from lymphocytes of 45 patients from sporadic ALS (29 males and 16 females), born between 1941 and 1985, were subjected to qPCR in order to study expression levels of *COL19A1, GSR, SNX10, MEF2C and NOGO A*. The levels found in every sample were related to survival values. Patients were classified in two groups (with less and more of 5 years of survival from symptoms onset) and the survival curves resulted between these 2 groups were significantly different on a Kaplan Meyer study.

Results: Preliminary results showed parallel correlations in *GSR, SNX10* and *NOGO A* with respect to longevity in both groups, while in the case of *COL19A1* the linear regression observed in the group of more of 5 years of survival from symptoms onset was coincident to previous results in the animal model and in muscle and lymphocytes samples from ALS patients.

Discussion and conclusions: These findings allowed us to develop a gene and protein expression biomarker algorithm to determine the predictive capacity although further serial samples are needed to confirm the relationship between the expression profile variation of the selected genes and survival of the patients.

Acknowledgements: This work was supported by grants PI10/00092, PI10/01787 and EC08/00049

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References:

1. Calvo AC, Manzano R, Atencia-Cibreiro G *et al* PLoS One 2012; 7:e32632.
2. Nachmany H, Wald S, Abekasis M *et al*. Disease Markers 2012; 32: 211-220.
3. Pradat PF, Bruneteau G, González de Aguilar JL, *et al*. Ann. Neurol. 2007; 62: 15-20.

SW14 QUANTIFICATION OF CFS CYTOKINES IN ALS BY A MULTIPLEXED BEAD-BASED IMMUNOASSAY

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Keywords: CFS, cytokine, flow cytometry

Background: Neuro-inflammation and production of pro-inflammatory cytokines have been involved in amyotrophic lateral sclerosis (ALS) pathogenesis.

Objectives: The aim of our work was trying to distinguish between control donors and ALS patients measuring cerebrospinal fluid (CFS) cytokines using a flow cytometry multiplex

Methods: We analyzed IL-17, INF-γ, TNF-α, IL-2, IL-4, IL-6 and IL-10 in CSF of 7 patients with probable or definite ALS according to El Escorial criteria (4 men and 3 women, 58.4±13.1 mean age, all of them with spinal onset) and 6 sex- and age-matched healthy controls (3 men and 3 women, 50±16 mean age) by a Cytometric Bead Array using antibody-coated beads and a FACS Calibur flow cytometer (Becton-Dickinson).

Results: INF-γ concentration was 6.89±0.21 (mean ± SD) pg/mL for controls and 6.69±0.31 pg/mL for ALS patients. TNFα mean concentration was 6.56±0.25 pg/mL for controls and 6.67±0.32 pg/mL for ALS patients. IL-2 mean concentration was 7.31 ± 0.16 pg/mL for controls and 7.28±0.13 for ALS patients. IL-4 mean concentration was 6.92±0.2 pg/mL for and 6.89±0.33 pg/mL for controls ALS patients. IL-6 mean concentration was 8.14±0.53 pg/mL for controls and 7.94±1.41 pg/mL for ALS patients. IL-10 mean concentration was 6.54±0.16 pg/mL for controls and 6.43±0.23 pg/mL for ALS patients. IL-17 mean concentration was 8.45±1.85 pg/mL for controls and 7.67±1.87 pg/mL for ALS patients.

Discussion and conclusions: We did not find significative differences between the measured cytokines in CFS of ALS patients and healthy donors maybe because of the limited number of samples used in this preliminary study. Increasing the number of samples from both ALS patients and healthy controls may lead to different conclusions, as it has been published that cytokines such as IL-12, IL-15, IL-17 and IL-23 were elevated in CFS of ALS patients in comparison with patients with non-inflammatory neurological disorders (1, 2). Healthy controls were not tested in these works and measurement method was ELISA.

References:

1. Rentzos M, Rombos A, Nikolaou C, et al, Interleukin-15 and interleukin-12 are elevated in serum and cerebrospinal fluid of patients with amyotrophic lateral sclerosis. Eur Neurol. 2010;63(5):285-90
2. Rentzos M, Rombos A, Nikolaou C, Interleukin-17 and interleukin-23 are elevated in serum and cerebrospinal fluid of patients with ALS: a reflection of Th17 cells activation? Acta Neurol Scand. 2010 Dec;122(6):425-9.

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SW15 BIOCHEMICAL AND CLINICAL MARKERS FOR MOTOR NEURON DISEASE SUBTYPES

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Keywords: cytokines, neuroinflammation, biomarkers

Background: Several pathogenetic hypotheses have been postulated for ALS/MND: recently neuroinflammation has been thought to play a key role.

Objectives: Our main aim is to find a correlation between many blood parameters (i.e. cytokines, growth factors, selectins, lymphocyte phenotypes, etc.) and clinical features; secondly, we intend to figure out specific markers correlated to ALS/MND subtypes.

Methods: Thirty patients with upper and/or lower motor neuron involvement were evaluated by means of the Medical Research Council (MRC) Scale, electromyography and motor evoked potentials, the ALS Functional Rating Scale (ALS-FRS), spirometry, and soft tissue analysis (STA). Clinical data were diachronically correlated to different blood parameters, during a 2-year follow-up period, using the statistical program *R*.

Results: In patients with first motor neuron involvement (MND1), a significant ($p<0.05$) positive correlation was found between total MRC scores and IL-1, IL-10, and CD-45RA levels; while TGF-β1, L-Selectin, VEGF, SOD-1, and CD-19 resulted negatively correlated with MRC ($p<0.05$). As regards patients with second motor neuron involvement (MND2), CD-25, CD-40 Receptor (CD-40R), CD-45RO, and Apo1-FAS showed a highly significant ($p<0.01$) positive correlation with MRC, whereas MMP-9, PDGF, MCP-1, CD-3 and CD-4 were reported to be negatively correlated ($p<0.01$). Interestingly, patients with both first and second motor neuron involvement (MND1+2) showed a different biochemical pattern, with CD-3 and CD-4 directly proportional to strength, while IL-6 Receptor (IL-6R), CD-44, CD-16/56, CD-19, CD-45RA, IGF-1, and MCP-1 were inversely correlated with it ($p<0.01$). Changing parameters in parallel with the strength loss include IL-1, IL-3, IL-6, TNFR-I, Apo1-FAS, VCAM-1, MMP-9, CD-3 and Vitamin B12 in MND1 patients; IL-6, TNF-α, ICAM-1, and CD-4 in MND2 patients; IL-6R, TNFR-II, P Selectin, MCP-1, ROS, CD-3, CD-4, CD-16/56, CD-25, CD-45RA, Vitamin B12 and folates in MND1+2 patients ($p<0.01$).

Total ALS-FRS scores were shown to be directly related to IL-8, CD-44, CD-16/56, CD45, CD-45RA, Glutathione Peroxidase activity, and Vitamin B12 ($p<0.01$), and inversely related to CD-45RO, CD-25, Glutathione Reductase activity, total anti-oxidant power, VEGF, CD-40R, ICAM-1, and IL-3 ($p<0.05$) in MND1 patients. It is proportional to IL-10, IFN-β , Apo1-FAS, CD-8 ($p<0.01$) and inversely proportional to MMP-9, VEGF, MCP-1, CD-16/56 in MND2 patients. In MND1+2 subjects, IL-6R, CD-44, IGF-1, CD-45RA, and Vitamin B12 had a negative correlation with ALS-FRS ($p<0.01$), while E-selectin and FGF had a positive one ($p<0.05$).

When compared to FEV1 values, many biochemical parameters showed significant ($p<0.01$) correlations in both mean values and their variations over time. Several blood parameters resulted significantly ($p<0.01$) correlated also to STA phase angle values: cytokines were different among MND1, MND2 and MND1-2 patients, and often corresponded with those that were significantly related to ALS-FRS and/or MRC scores.

Conclusion: Specific cytokine patterns for upper or lower motor neuron involvement were found, which could be useful in clinical and research settings.

SW16 P2 RECEPTOR EXPRESSION AND MODULATION OF THE ERMCC IN PERIPHERAL BLOOD MONONUCLEAR CELLS FROM PATIENTS WITH AMYOTROPHIC LATERAL SCLEROSIS

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Keywords: P2X, calcium homeostasis, PBMC

Background: In amyotrophic lateral sclerosis (ALS) adequate diagnostic and monitoring disease markers are desperately lacking, despite the severe nature of this progressive neurodegenerative disease. Peripheral blood mononuclear cells (PBMCs) which are involved in numerous diseases may resonate to both neurodegenerative and neuroinflammatory events. Ca^{2+} dysregulation plays a central role in the pathophysiology of ALS which is now thought to be a multi system disorder. In PBMCs, ER Ca^{2+} stores can interact with lysosome Ca^{2+} stores either directly

through activation of ryanodine receptors, or indirectly through release of ATP and engagement of P2X receptors, so that ALS related system pathology of the ERMCC may be detected in PBMCs.

Objectives: The aim of our study was to determine (i) P2X4R and P2X7R expression, and (ii) function, induced by direct P2X4 and indirect P2X7 receptor stimulation, as determined by cytosolic free Ca^{2+} influxes measurements and (iii) elucidate the role of P2XR activation in ER pathophysiological alterations of PBMCs from ALS patients.

Methods: We used PBMCs derived from ALS patients and age and sex matched healthy controls (HC). Cells were stimulated by exogenous ATP, an inflammatory stimulus (lipopolysaccharide, LPS), an inhibitor of the microsomal Ca^{2+}ATPase thapsigargin, and agonist for AMPA receptor (kainate). Calcium transients in individual monocytes were recorded by FURA-2 calcium imaging. P2XR expression was measured by immunohistochemistry and western blot.

Results: The immunostaining patterns of P2X4 showed decreased expression in ALS patients compared with controls. The same results were observed from western blot. In contrast, patients showed an elevated P2X7 expression. ER appeared swollen in monocytes from ALS patients. However, the volume level of mitochondria did not change. We observed cytosolic calcium load after exposing cells to drugs, The peak amplitude progressively increased in HC remained constant high after application with ATP in HCs but decayed faster in patients.

Conclusion: A growing body of evidence suggests that Ca^{2+}homeostasis dysregulation in ALS exist not only in motor neurons but also in immune cells. Purinergic signaling events regulate the activation and functional responses of monocytes and influence ER function. This may help to understand ALS pathophysiology on a systemic level and contribute as non-invasive surrogate marker for clinical trials in ALS.

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SW17 MODULATING ER CALCIUM UPTAKE WITHIN THE ERMCC IN ALS

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Keywords: SERCA, ERMCC, calcium

Background: Amyotrophic lateral sclerosis (ALS) is a deadly neurodegenerative disease that develops due to loss of motor neurons. Intracellular Ca^{2+} homeostasis is impaired and poses a key event in motor neuron degeneration and organelle dysfunction. We hypothesize that the ER-mitochondria Ca^{2+} cycle (ERMCC) might be center of the dysregulation. Ca^{2+} uptake into the ER by SERCAs (sarco (endo) plasmic reticulum Ca^{2+} ATPases) is essential for stable protein folding which is impaired in ALS. Selective motor neuron vulnerability may resonate in differential SERCA expression in motor neurons and non-motor neurons.

Objectives: (i) to investigate SERCA isoforms in vulnerable motor neurons at the molecular level in non-transgenic and G93A hSOD1 co-cultures, namely SERCA1 and SERCA2. (ii) to investigate the ability of SERCA activators to rescue motor neurons from dysregulation.

Methods: We used immunocytochemistry to study cellular localization and distribution of SERCA. mRNA-levels of SERCA1 and SERCA2 were monitored by qRT-PCR. Survival assays captured SERCA activator effects (6-Gingerol and Ochratoxin A) which were tested in motor neuron-enriched cultures for 12 h.

Results: So far, both isoforms are located in cell body of motor neurons, probably on the ER membrane. Subcellular localization of SERCA1 and SERCA2 and specific expression patterns of each isoform in non-motor neurons and motor neurons in both non transgenic and hSOD1 neurons will be presented. We expect to see a positive effect of SERCA activators on the survival of motor neurons.

Conclusion: The expression level of SERCA isoforms indicate a link between altered function and expression of SERCA within the ERMCC. Thus.

modulating SERCA may stabilize protein folding in the ER and thus slow disease progression in ALS.

References:

1.Grosskreutz J et al., 2010. Cell Calcium 47 165-174 10.1016/j.ceca.2009.12.002
2.Lautenschlager et al., 2013 Exp. Neurol. 13 166-167 10.1016/j.expneurol.2013.03.027
3.Chin et al., 2014 Am J Physiol Cell Physiol. 307 (11):C1031-8. 10.1152/ajpcell.00237.2013

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SW18 A NOVEL FUS KNOCK-IN MOUSE WITH FRAMESHIFT MUTATION TO INVESTIGATE THE PATHOBIOLOGY OF FUS-ALS

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Keywords: FUS, mouse, frameshift

Background: FUS is an ALS-causative gene that plays key roles in RNA metabolism (1, 2). We have modified the endogenous mouse *Fus* locus, humanizing the 3' end and introducing a human ALS mutation, 'FUS delta14'. The delta 14 mutation is a splice junction point mutation, that results in a missense change which deletes the FUS nuclear localization signal and produces a short string of aberrant amino acids at the carboxyl terminus - resulting in onset of ALS at the age of 20 years and disease duration of 22 months (3).

Objectives: This study is focused on characterising a novel FUS knock-in mouse model of ALS to examine the pathobiology of ALS. We are also investigating mutant FUS protein-specific function using the C-terminal frameshift nonsense peptide as a unique protein tag.

Methods and results: To define behavioural characteristics of our novel mouse model a large cohort (15 mice per sex per genotype) was assessed using a longitudinal phenotyping pipeline that measures both motor and cognitive function. A histopathology profile has been generated with cohorts of 10 mice per sex per genotype at four age time-points (2-, 6-, 9- and 12-months of age). To investigate the specific role of the mutant FUS protein we have developed a novel antibody that specifically recognises the frameshift nonsense peptide. This antibody was used to identify cellular localisation as well as protein-protein and protein-RNA interactions.

Conclusion: We have developed a novel mouse model of FUS-ALS by (a) modifying the endogenous *Fus* locus in mouse so that our modified gene is tightly regulated in a physiologically relevant manner and (b) choosing a human mutation that naturally creates a novel epitope, allowing for mutant-specific analysis and further filtering of whole transcriptome (and proteome) datasets. By combining these approaches we can best identify the changes from loss and gain of function of FUS that are most relevant to ALS and also increase our general understanding of FUS biology.

References:

1. Kwiatkowski TJ, Bosco DA, Leclerc AL, et al. Mutations in the FUS/TLS gene on chromosome 16 cause familial amyotrophic lateral sclerosis. Science. 2009;323(5918):1205-1208. doi:10.1126/science.1166066.
2. Lagier-Tourenne C, Cleveland DW. Rethinking ALS: the FUS about TDP-43. Cell. 2009;136(6):1001-1004. doi:10.1016/j.cell.2009.03.006.
3. DeJesus-Hernandez, M et al. (2010) De Novo Truncating FUS Gene Mutation as a Cause of Sporadic Amyotrophic Lateral Sclerosis. Human Mutation 31, E1377-E1389

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SW19 A NEW FUS-Δ14 MOUSE MODEL TO DISSECT THE PATHOBIOLOGY OF FUS-ALS

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Keywords: animal model, FUS, RNA metabolism

Background: The RNA-binding protein FUS is encoded by one of several ALS causative genes; it is involved in alternative splicing and regulating gene expression. FUS displays similarities to the RNA-binding protein TARDDP, and this suggests that disruption in mRNA metabolism may be a major contributor to ALS pathobiology (1, 2).

Objectives: This study focusses on characterising a novel FUS knock-in mouse model to examine the pathobiology of ALS. This model expresses a splicing mutation that has been shown to cause early-onset ALS in humans (3).

Methods: Pathological changes at the neuromuscular junction (NMJ) and changes in muscle fibre composition are an early manifestation of motor neuron dysfunction. Therefore, the tibialis anterior and extensor digitorum longus muscles will be dissected to perform NMJ innervation counts and muscle fibre typing in age-matched mice cohorts. To test for defects in splicing and gene expression regulation, adult cortices and spinal cord tissue from wild-type and heterozygous mice will be dissected and used to perform RNA sequencing.

Results: It has been suggested that the disease process progresses proximally from the synapse towards the cell bodies of motor neurons (4). The NMJ innervation counts and the muscle fibres might therefore help to identify pre-symptomatic pathogenic alterations. The RNA sequencing experiments may provide further insight into the biological processes disturbed by the expression of mutant FUS and may assist with the identification of potential therapeutic targets.

Conclusion: This novel model may provide a new resource in FUS-ALS research to investigate the molecular mechanisms that cause this fatal disease.

Acknowledgements: Funding is provided by the Medical Research Council (MRC) and the Motor Neurone Disease Association.

References:

1. Kwiatkowski TJ *et al.* Science 2009; 323: 1205-1208
2. Lagier-Tourenne C and Cleveland DW Cell 2009; 136: 1001-1004
3. DeJesus-Hernandez M *et al.* Hum. Mutat. 2010; 31: E1377-89
4. Fischer LR *et al.* Exp. Neurol. 2004; 185: 232-240

SW20 EXPLORING THE FUNCTIONAL PROPERTIES OF C9 AND SOD1 PATIENT SPECIFIC IPSC-DERIVED ASTROCYTES AND NEURONS

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Keywords: iPSC, ALS, astrocytes

Work using animal models have indicated electrophysiological phenotypes during development in both upper and lower motor neurons in the SOD1 variant of ALS. There is increasing evidence for an early transitory hyper-excitable phenotype in ALS, replaced by hypo-excitability at some point prior to disease onset in both upper and lower motor neurons. However, the mechanisms underlying electrophysiological phenotypes or how they relate to lower motor neuron loss are not yet understood. Disease phenotypes have also been identified in ALS astrocytes, and there is significant evidence suggesting the cell type is directly involved in the eventual death of lower motor neurons.

Induced pluripotent stem cell (iPSC) derived upper and lower motor neurons potentially provide powerful new models for studying ALS. Although there are some questions regarding the developmental stage iPSC-derived motor neurons represent, electrophysiological phenotypes have been observed. However, these iPSC-derived models have relied upon wild-type (WT) rodent isolated astrocytes to electrophysiologically mature the iPSC-derived motor neurons sufficiently to see significant levels of electrophysiological function. Therefore, this model system entirely lacks the influence of human ALS astrocytes on the functional development of ALS lower motor neurons. In our study, we are characterizing both iPSC-derived astrocytes and lower motor neurons from both C9 and SOD1 iPSC lines which have been derived from separate differentiation protocols to produce enriched populations of each cell type. We then utilize these enriched iPSC-derived astrocytes to enhance the

electrophysiological maturity of iPSC-derived motor neurons in a co-culture system. Further, this enables us to generate co-culture systems using WT, C9 and SOD1 iPSC-derived astrocytes and neurons. Therefore, in characterizing the astrocytes and neurons following co-culture we can ask hypotheses regarding the influence of astrocytes on neuronal development and disease phenotypes in ALS.

SW21 EXTENDING SURVIVAL IN THE MURINE MODEL OF ALS BY PROMOTING THE M2 MICROGLIAL STATE AND ENHANCING NEURONAL TROPHIC SUPPORT

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Keywords: infusion, mouse, SOD

Background: Multiple lines of research implicate both neurons and microglia in the progression of ALS. Neuronal trophic support can be achieved by directly increasing metabolic support to neurons or indirectly by decreasing harmful substances, such as inflammatory cytokines. Recent biomarker analyses from our laboratory indicate that ALS patients with a longer survival time have increased levels of trophic factors that promote the reparative M2 microglial phenotype, suggesting that microglial polarization is an important aspect of disease progression (1).

Objective: To determine if providing metabolic support *in vivo* to neurons and promoting the M2 microglial phenotype extends survival in the murine model of ALS.

Methods: At 70 days of age, mice with the $SOD1^{G93A}$ mutation underwent surgery to permit continuous infusion into the lateral ventricle; pumps were replaced at 100 days. There were a total of four groups: animals that received infusion of factors that promote M2 microglial polarity, animals that received infusion our trophic formulation based on cerebrospinal fluid, termed SCSF, animals that received infusion of phosphate buffered saline (PBS) and a No Surgery group. Disease onset was assessed by performance on the rotarod apparatus, and endpoint was determined by the inability of the animal to right itself after 30 seconds.

Results: Preliminary results suggest that infusion with SCSF or factors that promote M2 microglial polarity extends median survival rates in the murine model. As compared to No Surgery, SCSF increased survival by 7 days and infusion with M2 factors by 5.5 days. Infusion with PBS was of no benefit, as these mice had the same median survival rate as compared to the No Surgery mice in which no intervention to disease course was made.

Discussion and conclusions: Our data suggest that providing trophic factors via CSF infusion produces a meaningful extension of lifespan in the murine model. The data further indicate that the delivery of trophic factors is important and can not simply be infusion of a buffer solution that clears the CSF. Research is ongoing to optimize the composition of the SCSF.

Reference:

1. Su XW, et al., Biomarker-based predictive models for prognosis in amyotrophic lateral sclerosis. JAMA Neurol, 2013. 70(12): 1505-11.

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SW22 MOTOR NEURON DISEASE, GLUTAMATE EXCITOTOXICITY AND OXIDATIVE STRESS: A PILOT STUDY IN DOUBLE BLIND WITH A DIETARY SUPPLEMENT DONOR CURCUMIN

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Keywords: Curcumin, oxidative stress, glutamate

Oxidative stress involvement and glutamate excitotoxicity have been strongly hypothesized among the possible pathogenic mechanisms of motor neuron degeneration in amyotrophic lateral sclerosis (ALS). Various evidence in literature have shown a possible link between the two mechanisms. It is

assumed, in fact, that oxidative damage to the glutamate transporter EAAT2 may lead to the neurotransmitter extracellular compartment accumulation resulting in excitotoxic neuronal damage. The glial glutamate transporter EAAT2 plays a major role in glutamate clearance quickly removing glutamate from the synaptic cleft to prevent glutamate receptor overstimulation. Glutamate taken up by perisynaptic astrocytes is then converted to glutamine by glutamine synthetase. Recent studies have highlighted some interesting interactions of Curcuma Longa with intracellular redox balance, in particular Curcumin acts as a free radical scavenger and antioxidant, inhibiting lipid peroxidation. The purpose of this work is to evaluate, in ALS patients, the effect of oral treatment with Curcuma Longa (1500 mg) on parameters of oxidative stress (advanced oxidation protein products - AOPP, iron reducing ability of plasma - FRAP, and plasma thiols) and excitotoxicity (serum EAAT2 mRNA) also relating to an incremental aerobic exercise, through a double-blind placebo-controlled study, lasting 6 months to check the possible effect of Curcuma Longa on redox status and on the mechanisms of processing EAAT2 messenger and the subsequent processes of glutamate re-uptake. The study is now ongoing and preliminary results will be available by October.

References:

1. Lin CL, Kong Q, Cuny GD, Glicksman MA. Glutamate transporter EAAT2: a new target for the treatment of neurodegenerative diseases. Future Med Chem. 2012 Sep;4(13):1689-700.
2. Kim DS, Kim JY, Han Y. Curcuminoids in neurodegenerative diseases. Recent Pat CNS Drug Discov. 2012 Dec;7(3):184-204.
3. Barber SC, Shaw PJ. Oxidative stress in ALS: key role in motor neuron injury and therapeutic target. Free Radic Biol Med. 2010 Mar 1;48(5):629-41.

SW23 L-SERINE AS A TREATMENT FOR ALS

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Keywords: l-serine, Phase 1 trial, BMAA

Background: Guamanian ALS/parkinsonism/dementia complex (ALS/PDC) is probably caused by the dietary consumption by the Chamorros of the substituted amino acid, β-N-methylamino-L-alanine (BMAA), derived from cyanobacteria in the coralloid roots of the Cycad tree. BMAA is bioconcentrated in the brains of patients with ALS/PDC, ALS, Alzheimer's and Parkinson's diseases. BMAA chronic neurotoxicity produces pathological changes typically seen in ALS/PDC in a primate model. BMAA causes neurotoxicity by several different mechanisms, one of which is by misincorporation into neuroproteins via L-seryl tRNA synthetase, causing protein misfolding, cytoplasmic aggregates, and apoptosis. *In vitro* and preclinical *in vivo* studies suggest that L-serine blocks BMAA misincorporation.

Methods: We are completing a Phase 1 study of L-serine as a treatment for ALS. Twenty ALS patients will be enrolled into a 6-month double-blind randomized dose-ranging study of L-serine taken in doses ranging from 0.5 g to 15 g twice daily. Routine safety and efficacy measures are collected monthly. Plasma and CSF concentrations of L-serine and BMAA are determined at baseline and 6 months.

Results: To date 15 ALS patients have been enrolled. One patient withdrew because of stomach bloating. No other safety or tolerability issues have occurred. The study remains blinded but the slope of progression of the ALSFRS-R scale, an ALS functional measure, is about half that seen in comparable placebo-controlled trial patients with the same entry criteria. Full results including efficacy measures, plasma, and CSF concentrations will be presented.

Conclusions: Preclinical studies suggest that L-serine may be of value in treating/preventing BMAA toxicity. A phase 2 trial in ALS is being planned.

Acknowledgements: Supported by the Fellows and Directors of the Institute for Ethnomedicine.

SW24 THE CANALS STUDY: A RANDOMIZED, DOUBLE-BLIND, PLACEBO-CONTROLLED, MULTICENTRE STUDY TO ASSESS THE EFFICACY ON SPASTICITY SYMPTOMS OF A CANNABIS SATIVA EXTRACT IN MOTOR NEURON DISEASE PATIENTS

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Keywords: therapy, cannabinoids, spasticity

Background: Spasticity is a one of the major determinant of functional loss and decline in quality of life in ALS and other motor neuron disease (MND) patients. Available anti spastic therapy results are often unsatisfactory, moreover, there are no randomized trials that evaluate anti spastic drugs efficacy in ALS patients. This means that spasticity treatments in ALS are not evidence supported. There is a strong rationale to propose a cannabis derived drug to treat ALS spasticity. Cannabinoid receptor have been found in brain and spinal cord and exogenous agonists for cannabinoid receptor show inhibitory properties. In recent years, several clinical trials have tested the efficacy of cannabis on spasticity in multiple sclerosis. A few reports signal an unlicensed cannabinoids use of several ALS patients to manage spasticity. Moreover, cannabinoids have central anti-inflammatory, anti-glutamatergic and anti-oxidant actions. Cannabinoids ability to target multiple neurotoxic mechanisms raises them as ALS therapeutic candidates. Recent studies on animal models of ALS have shown cannabinoids efficacy in improve survival and reduces movement dysfunctions.

Objectives: The study's primary aim is to evaluate the safety, tolerability and efficacy of a Cannabis Sativa extract medium-term treatment to improve spasticity in ALS and MND patients.

Methods: We designed a Randomized, Double-Blind, Placebo-Controlled, parallel groups, Multicentre Study to Assess the Efficacy on Spasticity Symptoms of a Cannabis Sativa Extract in MND Patients (Phase A, total duration: 6 weeks). The study will be followed by an open-label, follow-up phase in which all patients will take the active drug (Phase B). 60 consecutive patients fulfilling specific inclusion and exclusion criteria will be recruited, after informed consent. Subjects included will be randomized and double blinded allocated to receive a cannabis extract oral spray or placebo. Primary end-point will be improvement in the modified 5- points modified Ashworth scale. Secondary End-points will be spasticity, spasm frequency and sleep disruption (0 - 10 NRS score); Function: walking ability, functional scores (ALS-FRS-R); pain (0 - 10 NRS score).

Results: Patient recruitment and follow-up have just been concluded at the time of abstract submission. The results of study data analysis will be available and presented at the meeting.

Discussion and conclusions: Cannabinoids may represent a valuable option for spasticity treatment in ALS patients. Moreover, it may have an additional beneficial effect on ALS patients quality of life, as it may exert an analgesic effect. Finally, demonstration of cannabis based drug safety and tolerability may stimulate further studies aimed at evaluating a potential cannabis neuroprotective effects on ALS.

Acknowledgements: This study has been funded by Fondazione Italiana di Ricerca per la Sclerosi Laterale Amiotrofica (AriSLA). GW / Almirall kindly provided study drug /placebo.

SW25 LUNASIN VIRTUAL TRIAL: A NOVEL PATIENT-CENTRIC DESIGN

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Keywords: Lunasin, trial, enrollment

Background: Traditional ALS trials have several challenges. Hypotheses often come from observations made in animal models of ALS; how well these predict sporadic human ALS remains unclear. Dosing is difficult to gauge due to lack of pharmacodynamic

biomarkers. Enrollment may be slow, and retention poor due to narrow inclusion criteria, patient and family desire to avoid placebo treatment, frustration with the length of time it takes to get trial results, and study burdens including frequent trips to the study site. Here we report on the design of a novel pilot trial, which we hope will be able to overcome these challenges.

Methods: The rationale for this came from a patient with PMA, who experienced validated, clinically significant improvements on a Lunasin supplement regimen. We will test this same regimen in 50 more PALS, looking for improvements in ALSFRS-R scores over 12 months. Because we are looking for such a large signal, our trial can be widely inclusive. Lunasin reportedly alters histone acetylation, a measureable mechanism targeted in at least one previous ALS trial (sodium phenylbutyrate). Thus, we will employ a pharmacodynamic marker: histone acetylation before and during Lunasin treatment. We will utilize historical controls. Most of the visits in this trial will be virtual; we will teach PALS to measure ALSFR-S, weight, perceived efficacy, and side effects and to record these on the website PatientsLikeMe. Results from this study will be available in real time. These features should boost enrollment and retention.

Results: Our novel design will allow us to test the following hypotheses:

1. Lunasin decreases the rate of ALSFRS-R progression by 50% relative to historical controls.
2. Lunasin increases the frequency of ALS reversals (defined by an improvement of 4 or more points in the ALSFRS-R over the course of 1 year) to at least 2%.
3. Lunasin alters histone acetylation in PALS.
4. Participants can accurately measure their ALSFRS-R score and record it on PatientsLikeMe.
5. Participants can accurately measure their own weight and record it on PatientsLikeMe.
6. The novel features of this pilot trial will be associated with improved participant enrollment compared to prior more traditional ALS trials where this is 2 participants per site per month.
7. The novel features of this pilot trial will be associated with improved participant retention compared to prior more traditional ALS trials where the drop out rate is 22%.

Conclusions: The Lunasin Virtual Trial will employ a number of patient-centric features designed to circumvent problems encountered in more traditional studies. This will open in the early summer of 2015 and preliminary results including enrollment rate and histone acetylation should be available by the date of the Symposium.

THEME CW: CLINICAL WORK IN PROGRESS AND CARE PRACTICE

CW1 MRI-HISTOLOGY CORRELATES OF CORTICAL AND WHITE MATTER CHANGES IN POST-MORTEM MND/FTD BRAIN

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Keywords: MRI-histology correlation, white matter tracts, post-mortem MRI

Background: Novel magnetic resonance imaging (MRI) techniques enable the identification of significant cortical and white matter changes in patients with motor neuron disease (MND) and/or frontotemporal dementia (FTD). In MND, alterations are primarily observed in the primary motor cortex (M1), corpus callosum and corticospinal tract (CST) (1). In FTD, white matter involvement may be more extensive than detectable cortical atrophy, and this may represent the early phase of FTD (2). However, the microstructural and molecular pathologic correlates of these MRI measures remain poorly defined and are ideally demonstrated histopathologically. Therefore, MRI-histology correlation analysis in post-mortem MND/FTD tissue would be a crucial platform for characterising specific MRI biomarkers.

Objectives: To histologically measure differential microstructural and molecular changes in cortical and deep white matter regions relevant to disease, and correlate them with specific MRI signal changes in post-mortem MND/FTD brains.

Methods: Post-mortem brains (10 MND/FTD and 5 controls) will undergo enhanced 7T MRI imaging for measures that are sensitive to microstructural properties including structural scans (cortical thickness), relaxography (myelin content), susceptibility-based contrast (iron), magnetisation transfer (myelin and gliosis) and high-resolution diffusion weighted steady-state free precession (DW-SSFP; for more specific measures of white matter) (3). Following scanning, the M1, medial prefrontal gyrus, CST, corpus callosum, cingulum and fornix will be sampled, guided by anatomical landmarks and tractography. The relative burden of pathology in regions of interest and disease progression will be quantified in sections processed for TDP-43 and p62; Iba1/CD68 (microglia/macrophage); Perl's and Turnbull's iron; luxol fast blue and myelin protein (myelin integrity); and neurofilament content. These measures will be analysed in relation to specific MRI parameters from matched regions of interest.

Results: We present our study protocol (6 MND brains, 1 spinal cord), post-mortem tractography and initial MRI/pathology data of CST degeneration.

Conclusion: We believe our approach will help to define the microstructural and molecular pathologic correlates of novel MRI techniques that are currently applied *in vivo*.

References:

1. Turner MR, Verstraete E, Curr Neurol Neurosci Rep 2015; 15:569.
2. Mahoney CJ, Ridgway GR, Malone IB, *et al* Hum Brain Mapp 2014; 35:4163-4179.
3. Foxley S, Jbabdi S, Clare S, *et al* Neuroimage 2014; 102:579-589.

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CW2 AUTOMATED DETECTION OF FASCICULATIONS FROM B-MODE ULTRASOUND IMAGES FOR MOTOR NEURONE DISEASE DIAGNOSIS

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Keywords: ultrasound, fasciculations, automated

Background: Electromyography (EMG) is the standard diagnostic technique for the detection of fasciculations (twitches) that present in neuromuscular disorders such as MND (1). Ultrasound (US) imaging may provide a more sensitive alternative to EMG for detection of fasciculations (2), however computational techniques have not been used to objectively identify involuntary tissue displacements in comparison to electrophysiological activity.

Objective: To determine whether fasciculations may be automatically detected in b-mode US images using a computer vision based foreground detection approach.

Method: US images of medial gastrocnemius (MG) and biceps brachii (BB) were collected from: Young (18-35 years, *N*=15) and older (50+ years, *N*=9) healthy adults and MND patients (*N*=5). EMG was simultaneously collected from five participants in each group. Two forms of analysis were completed, comparing computer vision techniques to fascicluations identified through: i) manual identifications (IDs) in collected US images; and ii) EMG data. Computational identification of fasciculations was completed using a foreground detection approach which models pixels as a mixture of gaussians, enabling objects of interest to be differentiated from the background image. 500 frames from each video were used to train the model, with 2300 frames used as test data. Receiver Operating Characteristic (ROC) curves were used to compare IDs and EMG signals to results, with the area under the curve used to indicate agreement levels between signals.

Results: Computational approach/manual ID comparison: 18 - 35 (*N*=15); MG - agreement of 94.29% from 213 Ids; BB - agreement of 93.48% from 38 Ids; 50+ (*N*=9); MG - agreement of 94.41% from 81 Ids; BB - agreement of 91.75% from 21 Ids; MND (*N*=5); MG - agreement of 90.96% from 133 Ids; BB - agreement of 91.60% from 141 Ids. Computational approach/EMG comparison: 18 - 35 (*N*=4); MG - agreement of 89.71% from 44 fasciculations; BB - agreement of 84.35% from 95 fasciculations; 50+ (*N*=5); MG - agreement of 86.59% from 89 fasciculations; BB - agreement of 81.11% from 26 fasciculations; MND (*N*=5); MG - agreement of 82.47% from 112 fasciculations; BB - agreement of 83.75% from 214 fasciculations.

Discussion and conclusion: Computational techniques show promise for automated, objective identification of involuntary activation. The automated technique when compared to IDs, showed very high levels of agreement throughout all test groups and the EMG comparison yielded good agreement. Differences in agreement levels may be due to complexity of EMG signals ,making it harder to extract fasciculation potentials. In addition, the EMG detection volume is also much smaller than the area assessed by US.

References:

1. de Carvalho M, Dengler R, Eisen A *et al* Clinical Neurophysiology; 119(3):497-503
2. Walker F O, Donofrio P D, Harpold G J *et al* Muscle & Nerve 1990; 13(1):33-39

CW3 NERVE CONDUCTION STUDIES IN 154 CASES OF AMYOTROPHIC LATERAL SCLEROSIS

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Keywords: action potentials, nerve conduction

Background: Nerve conduction studies are a test which is non-invasive and convenient, may potentially reflect the disease progression of amyotrophic lateral sclerosis (ALS). Here, we explored the evaluating effect of nerve conduction studies in ALS.

Objective: To analyze the features of nerve conduction in patients with ALS, and explore the correlation between compound muscle action potential (CMAP) amplitude and disease duration and revised Amyotrophic Lateral Sclerosis Functional Rating Scale (ALSFRS-R).

Methods: Standard motor and sensory nerve conduction were performed in 154 patients with ALS. Neurophysiological tests included CMAP amplitude, distal motor latency (DML) and/or motor and sensory conduction velocity. Explore the correlation between CMAP amplitude and disease duration and ALSFRS-R.

Results: A majority of patients presented motor nerve conduction abnormalities. Decreased MCV, prolonged DML, decreased CMAP amplitude and

absence of CMAP were found in 27.50%, 81.42%, 53.98% and 15.00% patients respectively. A small proportion of patients were abnormal in sensory nerve conduction. Decreased SCV, decreased SNAP amplitude and absence of SNAP were found in 2.73%, 1.82% and 1.22% patients respectively. There was significant positive correlation between median nerve and ulnar nerve CMAP amplitude and ALSFRS-
R（r=0.273，p=0.016；r=0.357，p=0.001）.

Discussion: Nerve conduction studies has clear potential to provide diagnosis that may be sensitive to both prognosis and disease assessment in ALS. Prolonged DML is accepted to reflect the degree of degeneration of axons in ALS, which provide evidence for the early-stage abnormalities of motor nerve conduction. There was significant positive correlation between median nerve and ulnar nerve CMAP amplitude and ALSFRS-R, which suggest CMAP amplitude of median nerve and ulnar nerve might be of certain clinical value in estimating the disease severity. However, this must be explored further in larger studies with longer follow up and multivariate analysis.

Conclusion: This study revealed motor nerve conduction were abnormal in a majority of ALS patients and prolonged DML is the most common one, abnormal sensory nerve conduction were found in only a few ALS patients. CMAP amplitude of median nerve and ulnar nerve might be of certain clinical value in estimating the disease severity of ALS.

CW4 PAIN AND ALS: A SYNERGISTIC DICHOTOMY?

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Keywords: pain, quality of life, targeted pain treatment

Background: Pain is an often understated and neglected symptom in ALS, in fact often cited as “rare”, as the pathophysiology of the disease does not have sensory involvement. However, pain is a commonly heard complaint of PALS which has been poorly studied without established targeted treatment guidelines. A negative effect on quality of life (QoL) might also be expected in ALS patients with pain when not managed correctly.

Objective: The aim of this study is to evaluate the prevalence of pain in ALS patients, including etiology, and treatment modalities used, as well as effects on quality of life.

Methods: 50 patients with ALS will be interviewed during routine clinical visits to assess for presence of pain. Those with reports of pain will then be stratified by etiology of pain (i.e. radicular, compression, cramps, neuropathic, and unknown). Data will be collected with the use of the short form brief pain inventory (BPI) along with information regarding various techniques to manage pain, including medication, PT, etc, as well as respective efficacy.

Results: Pending data collection.

Discussion: Our study aims to show that pain is a relatively frequent symptom with an important impact on PAL’s quality of life, which requires treatment. This data can also be used to educate clinicians and patients to promote better targeted multidisciplinary management of ALS symptoms and a better quality of life.

References:

1. Hanisch et al. Characteristics of pain in ALS. Brain Behav. March 2015, 5(3).
2. Pizzementi et al. Depression, pain and quality of life in in patients with ALS: a cross sectional study. Functional Neurology, 2013 April-May 28(2): 115-9.
3. Wallace et al. The evaluation of pain in ALS: A case contolled observational study. ALS FTD. 2014 Dec; 15(7-8): 520-7.

CW5 BENEFITS OF SCAPULAR MOBILIZATION FOR IMPROVING RANGE OF MOTION AND DECREASE PAIN OF THE AMYOTROPHIC LATERAL SCLEROSIS SHOULDER

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Keywords: painful shoulder, treatment, prevention

Background: Describe the benefits of utilizing scapular mobilization for improving range of motion

(ROM) of the affected Upper Extremity in patients with Amyotrophic Lateral Sclerosis.

Introduction: Persons diagnosed with ALS at an academic medical center are referred to a multidisciplinary clinic . The Occupational Therapist evaluates and treats patients in the ALS clinic who have developed a painful shoulder and or limited Passive Range of Motion (PROM) due to profound weakness. During the evaluation and treatment process in the ALS clinic, pain reduction has been documented in the shoulder following scapular mobilization. In addition, increases in ROM of the affected limb after gentle scapular mobilization of an average of 20 degrees forward flexion and abduction have been documented in multiple cases.

Objectives: The primary objective of this case series is to increase awareness of the potential benefits of scapular mobilization in the treatment of the affected limb in patients with ALS. The secondary objective is to develop educational materials for the clinician and caregivers in the management of the painful shoulder in individuals affected by ALS.

Methods: As part of the Occupational therapy evaluation in the multidisciplinary ALS clinic, the patient receives an assessment of the Active Range of motion (AROM) and PROM of the upper extremity through goniometric measurement. The shoulder pain is assessed using a 10 point visual analog scale.

For patients with decreased shoulder ROM and or painful shoulder ROM, the scapula is assessed for mobility. If mobility is limited, scapular mobilization is performed. Goniometric ROM was tested and Visual Analog Scale evaluated again following the scapular mobilization and in 6/6 cases the ROM improved by at least 20 degrees with decrease in pain experienced by the patient.

Conclusion: The use of Manual therapy, scapular mobilization to improve scapulohumeral rhythm of the affected Upper extremity in individuals affected by ALS is a valuable tool that in the cases discussed in this report has shown significant improvement in the painful shoulder, an unfortunate complication of upper and lower motor neuron weakness in ALS. There is a need to improve the standard of care for the patient suffering from ALS, increasing awareness of better techniques to address the specific needs of the affected upper extremity to decrease complications and improve quality of life.

Implications: The greatest challenge as a clinician treating patients with ALS is to find treatment techniques and approaches that improve functional status and quality of life. Avoidance of pain and of complications such as a painful shoulder must be carefully considered.

The use of scapular mobilization has proven to be beneficial in the cases presented. Further investigation will be the next step in assessing the benefits and optimal application in ALS care.

CW6 ENGAGEMENT IN PURPOSEFUL OCCUPATION COMPRISING AEROBIC ACTIVITY AND MUSCULAR RESISTANCE HAS POTENTIAL TO IMPROVE FUNCTION FOR PEOPLE WITH MND

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Keywords: rehabilitation, exercise, well-being

Background: At 2014 International Symposium the author presented "To Do or not to Do; The Conundrum for Therapists in Advising Level of Doing for MND" which produced lively discussion between therapists. The debate challenged whether therapy intervention should be within a rehabilitation or compensational framework. This study to explore effectiveness of rehabilitation with MND has received fellowship funding from Welsh Research Network and is now in 3rd year of study.

Objectives: 1.Identify effect of aerobic activity and muscular resistance on daily functioning for people with MND; 2. Explore patients experiences of rehabilitative approach.

Method: A case based approach with convenience sample of 6 people living in Wales diagnosed with MND, presenting with mild/moderate symptoms. The 6 participants receive 6-week rehabilitative intervention consisting of weekly occupational therapy sessions incorporating functional activity to moderate aerobic capacity and moderate muscular resistance. The Canadian Occupational Performance Measure (COPM) (1) assists tailoring intervention and as an outcome measure (initial assessment, immediate post-intervention and follow-up post 3 months of intervention). Measurement is both performance and satisfaction of performance within client identified functioning. Qualitative data is obtained through semi-structured interviews at 3-time points (initial assessment, immediate post-

intervention and 3 month follow-up). The interviews focus upon the participant's perception of their change in performance and satisfaction level, subjective accounts of changes in health and quality of life. An inductive approach to data analysis is utilised for the qualitative data and the interpretive strategy is based on adaptation of the constant comparative method. The quantitative data are analysed through repeated measures anova.

Results: At time of submitting abstract, 3 of the 6 case study interventions have been completed. Results of 3 case studies to date demonstrate an improvement in outcome measure for both performance and participants satisfaction with performance. Participants convey positive well being from challenging their system aerobically and within muscular resistance within an empowering approach.

Discussion and conclusions: Initial findings show that regular aerobic and resistance exercise can maintain function in mild/moderate presentations of MND. This confers with previous studies within MND (2). There is anecdotal experience that people with MND who "push themselves" to engage in activity achieve a plateau in symptoms whilst those with occupational deprivation have a more marked decline. Whilst the study is not yet complete there are emerging results which suggest a rehabilitative approach to therapy intervention is positive in terms of functional outcomes. By poster presentation all case study interventions will have been completed.

References:

1. Almeida et al (2012) Exercise and Amyotrophic Lateral Sclerosis. Neurological Sciences. 33 (1) 9-15
2. Law et al (2000) Canadian Occupational Performance Measure. CAOT Publications.

CW7 VITAL CAPACITY- HIGHLY VARIABLE IN PATIENTS WITH BULBAR WEAKNESS?

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Keywords: vital capacity, variability, bulbar weakness

Background: Median decline in forced vital capacity (FVC) is reported to be 2.0% per month in amyotrophic lateral sclerosis (ALS) patients. In clinical settings, we notice test-retest variability in breathing tests. Contributing factors of the variability have not been clearly identified.

Objectives: We analyzed test-retest variability of VC in patients with ALS, using the longitudinal records of slow vital capacity (SVC) in the Clinical Trial of Ceftriaxone.

Methods: In the Clinical Trial of Ceftriaxone in Subjects with ALS, 513 subjects were enrolled at 70 centers in US and Canada. We evaluated the variability of SVC using the database of the Clinical Trial of Ceftriaxone as follows. First, test-retest SVC changes at the same visit within each subject were analyzed. Within 3 measurements of vital capacity, we evaluated incidences of >10% test-retest variability in SVC within each subject. We also searched factors that contribute to this variability including ALSFRS-R bulbar subscore, ALSFRS-R respiratory sub-score, SVC<60, and time of the day. Second, prognostic potential of SVC variability at the screening visit was analyzed for the disease progression. Multiple evaluators' effect to SVC variability was also assessed.

Results: Our preliminary results showed that ALSFRS-R bulbar subscore strongly correlated with SVC variability ($p < 0.0001$). Other contributing factors include SVC < 60% and ALSFRS-R respiratory subscore. High test-retest variability (>10%) was seen in the patients with bulbar weakness at screening visit (15.2% of severe bulbar weakness patients had high SVC variability, compared with 5.5% of mild bulbar weakness patients). This trend became particularly noticeable at the week 48 of the clinical trial (32.7% of severe bulbar weakness patients had high SVC variability, compared with 8% of mild bulbar weakness patients). SVC variability was prognostic for total ALSFRS-R decline ($p < 0.0001$). Multiple evaluators' effect was not significant.

Discussion and conclusions: Test-retest variability of FVC is associated with bulbar weakness and prognostic for functional decline in the patients with ALS.

References:

Cudkowicz ME, Titus S, Kearney M *et al* Lancet Neurol 2014; 13:1083-1091
Traynor BJ, Zhang H, Shefner JM *et al* Neurology 2004; 63:1933-1935

CW8 DIURNAL INTERMITTENT ABDOMINAL PRESSURE VENTILATION (IAPV) WITH PNEUMOBELT IN AN ALS TRACHEOSTOMIZED PATIENT: A CASE REPORT

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Keywords: mechanical ventilation, rehabilitation, quality of life

Background: Pneumobelt consists of an elastic inflatable bladder incorporated within a corset surrounding the abdomen. With bladder inflation by a ventilator, the abdominal content and diaphragm move upward, assisting expiration. With bladder deflation, inspiration occurs passively.To our knowledge, this is the first report showing Pneumobelt use in tracheostomized Amiotrophic Lateral Sclerosis (ALS) patients.

Objectives: To assess the effects of Pneumobelt in a 49 year old, tracheostomized, tetraplegic man with spinal onset ALS, who required 24 h invasive ventilation support on admission at our department.

Methods: We optimized both invasive ventilation by a pressure single tube mode with a leak valve and secretion clearance by cough machine. To permit speaking, we introduced diurnal tracheal open ventilation (TOV), but with poor patient's tolerance because of trouble in air, secretions and saliva management. Thus, we introduced diurnal Pneumobelt associated to a tracheostomy speaking valve.

Results: Pneumobelt permitted an efficient diurnal ventilatory pattern, good pulmonary gas exchange, optimal speech, without dyspnea and with a significant reduction of secretion management's need. At present, our patient maintains IAPV for 3-4 h, while he is on wheelchair, with great compliance to treatment. He feels his breathing is "normal, as before tracheostomy".

Conclusions: Our case report suggests that Pneumobelt can be a safe and effective method of daytime ventilation, and, improving speech and secretion management, it permits a better quality of life even in tracheostomized ALS spinal patients.

CW9 OUTCOME STUDIES OF DIAPHRAGM PACING IN ALS

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Keywords: diaphragm pacing system,

Background: Diaphragm pacing (DP) is FDA approved for the treatment of hypoventilation in ALS and appears to be reasonably safe in carefully selected patients. There are no published controlled studies regarding the efficacy of DP in patients with ALS, although such a study is underway.

Objectives: Examine patient outcomes following DP surgery.

Methods: Patient criteria for DP surgery included negative inspiratory force (NIF) < -60 cm of H_2O, forced vital capacity (FVC) > 45% of predicted (less if bulbar symptoms present), patient stated goals of life extension through treatments, sniff fluoroscopy of the chest or phrenic nerve stimulation to assess diaphragm movement; and arterial blood gas measurements demonstrating hypercarbia (pCO2 > 45). The following data were collected for all patients undergoing DP surgery at our institution: forced vital capacity (FVC) and ALS Functional Rating Scale-R (ALSFRSR) scores every three months, time from ALS symptom onset to death or tracheostomy, monthly patient reports of DP usage, and of work of breathing, dyspnea, voice volume, energy, sleep, and secretion management. Descriptive statistics, Spearman's correlation and Wilcoxon rank sum tests were performed. The study received Institutional Review Board approval.

Results: To date, 11 patients have undergone DP surgery; 7 men and 4 women, mean age 54.5 years at ALS symptom onset; mean disease duration prior to surgery 27.6±14.13 months. In the 9 months prior to surgery, the mean decrease per month was 1.5% for FVC and 1.0 points for ALSFRSR. Patients have been followed for an average of 5.1 months post-DP surgery (range 1 - 12 months). At one month post-surgery, all patients paced 8 hours per day. Post-surgery, the mean decrease per month for all 10 patients with follow-up data was 3.4% for FVC and 1.1 points for ALSFRSR. Three patients died (4, 48 and 169 days post-operatively) and 1 patient required

tracheostomy/mechanical ventilation 3 days after DP surgery. Gender, disease duration, site of onset, and rate of change in FVC prior to surgery were not related to post surgery changes in FVC or ALSFRSR. Older patients had greater declines in FVC post surgery (r= -0.53). One patient reported improvements in sleep quality with pacing. All other patients reported no change in symptoms following DP surgery.

Discussion: This small non-blinded non-randomized study did not identify any disease characteristics beyond age that might contribute to a more rapid disease decline after surgery. Patient reported outcomes demonstrated that DP did not impact ALS symptoms. Conclusions regarding the efficacy of DP in ALS patients await larger, randomized studies.

CW10 UNIFORM METHODS FOR CLINICAL DATA COLLECTION: AN EPIC APPROACH FOR US ALS CENTERS

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Keywords: data, EPIC, prognosis

Background: In 2012, we surveyed US ALS centers and found that the majority use Epic as their institutional Electronic Health Record (EHR). Epic is one of the leading EHR vendors in the United States. Its platform allows designing, creating and deploying custom forms that can be shared across institutions. Our team has worked with Epic to design 14 ALS-specific forms that enable the collection of a variety of standard data within the workflow of clinic. These were completed in late June and the initial site installations were completed at CPMC and University of Minnesota in July. In the current phase, we will develop infrastructure and standard procedures for data aggregation across multiple sites. This will lead to uniform methods for collecting, capturing and extracting clinical information from all PALS seen across different ALS clinics.

Methods: The Epic ALS data platform captures information (date of onset, progression, longitudinal FRS and PFTs, genetic testing information, family history, labs, examination, riluzole use, etc.) within clinic workflow on every patient seen in clinic. The two pilot sites, in collaboration Epic, work with local legal IRBs, regulatory, and IT teams to develop standard processes operating procedures for collecting data for information from PALS . Standardized data elements will be exported from each site and sent to the central repository - NeuroBANK™ - under the umbrella of an already approved ALS Natural History protocol.

We will develop further plans for methodology, regulatory controls, data sharing, governance agreements and recommendations alongside Epic.The project team will produce specific guidelines that facilitate development, deployment and utilization of the ALS-specific forms and for the data extraction across the Epic EHR system.

Results: Fourteen ALS forms were released in late June for distribution to all Epic installations. An agreement to share data was also put in place. We have begun working with local IT teams on automated data export processes. We continue to work with Epic to update forms, create new forms, and establish wider governance. We anticipate beginning the initial stages of data capture and comparisons of overall data between our clinics in the late fall.

Conclusions: We have taken the initial steps for creating what could potentially become the largest all-inclusive dataset of bedside information from a PALS population attending ALS clinics, from the first visit onwards, without pre-conceived time frames or narrow eligibility criteria. We think this prospective approach to data will ultimately support projects in prognostication, epidemiology, best practices, comparative effectiveness, and utilization of resources. Ideally, biomarker and even interventional studies will see the value of having access to pre-existing prospectively obtained longitudinal with lead-in clinical data. The development of novel disease progression and survival models could also serve as the basis for future clinic-based trials.

CW11 PHYSICAL AND COGNITIVE FITNESS IN YOUNG ADULT MALES AND RISK OF AMYOTROPHIC LATERAL SCLEROSIS AT EARLY AGE

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Keywords: physical fitness, cognitive fitness

Objective: To explore the influence of cognitive and physical fitness during early adulthood on the risk of amyotrophic lateral sclerosis (ALS).

Methods: Data on physical fitness, body mass index (BMI), intelligence quotient (IQ) and stress resilience at age 18-20 were collected from 1,736,060 Swedish men, aged 18-20 at conscription examination during 1968-2009. Subsequent ALS diagnoses were identified through the Swedish Patient Register. Hazard ratios (HRs) and corresponding 95% confidence intervals (CIs) derived from Cox models were used to assess the associations of BMI, IQ, and stress resilience with ALS. The influence of physical fitness was estimated using flexible parametric models.

Results: We identified 239 ALS cases during follow-up. Mean age at diagnosis was 45 years. Five-unit increase of BMI was associated with a 23% lower risk of ALS (95 % CI 1%-40%). The time-varying association between physical fitness and ALS risk was positive and significant until age 40 (HR 1.55, 95% CI 1.03-2.33). IQ was not associated with ALS risk (HR 1.03, 95% CI 0.96-1.11), whereas higher stress resilience tended to be associated with a lower risk of ALS (HR 0.93, 95% CI 0.85-1.00).

Conclusions: BMI, physical fitness, and stress resilience, but not IQ, in early adulthood might be associated with a reduced risk of ALS. Further research is needed to assess whether ALS patients demonstrate lower stress resilience than average, both pre-symptomatically and clinically.

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CW12 THE RELATIONSHIP BETWEEN COGNITION AND DISEASE STAGING IN ALS

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Keywords: disease staging, cognition and behaviour status, screening

Background: The Edinburgh Cognitive and Behavioural ALS screen (ECAS) is a brief multi-domain assessment measuring cognition and behaviour in ALS, while accounting for physical disability (1). However, the relationship between cognition, behaviour and disease progression has yet to be confirmed. A disease staging system has been proposed in which patients' stage depends on the number of regions involved, and respiratory/nutritional intervention, with each stage marking a percentage disease course (2). Unfortunately, previous research has failed to fully incorporate extra-motor changes into a formal measure of staging.

Objectives: To determine the relationship between cognitive and behaviour change, and disease staging.

Methods: In total 160 incident patients will be recruited from three centres (Edinburgh, Dublin, London), in addition to 100 healthy controls. Participants will be seen every four months for four occasions. Participants will be administered a battery of tests including a staging system (2), a measure of cognition and behaviour (ECAS), the hospital anxiety and depression scale, and the ALS-FRS-R.

Results: Data have been collected on 74 patients from Edinburgh and Dublin for Phase 1 (preliminary analyses). Patients were stratified based on their

disease stage (Stage 1-4), with 28%, 28%, 12%, and 31% of patients impaired in each stage respectively. Evidence of cognitive impairment was found in language, executive functioning, verbal fluency, and memory in all stages. For visuospatial functioning, only patients in Stages 1 and 4 demonstrated impairments. Behavioural data were available for 69 patients, with 17%, 40% 22% and 43% of patients demonstrating behavioural abnormality in each respective stage. Kruskal-Wallis and Jonckheere-Terpstra tests were performed to determine whether cognitive profiles differed based on stage, with respect to stage order. Only executive functioning significantly differed between stages ($H(3)=8.86$, $p = 0.031$ and $J = 750$, $z = -2.18$, $p = 0.029$, $r = -0.25$). Pairwise comparisons with adjusted p-values showed a significant difference between Stages 2 and 4 ($p = .030$, $r = 0.418$).

Discussion and conclusions: Initial evidence suggests that the profile of cognition and behaviour in patients with ALS is present throughout all stages of the disease. However, executive dysfunction is highest in Stage 4. Decline in executive functions may impact on patient management, and patients' engagement with interventions.

References:

1. Abrahams S, Newton J, Niven E, *et al* (2014). Screening for cognition and behaviour changes in ALS. Amyotrophic Lateral Sclerosis and Frontotemporal Degeneration, 15(1-2), 9-14.
2. Roche JC, Rojas-Garcia R, Scott KM, *et al* (2012). A proposed staging system for amyotrophic lateral sclerosis. *Brain, 135*(3), 847-852.

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CW13 A COGNITIVE SCREENING TOOL FOR ALS PATIENTS: THE ALS-FTD-COG, PRELIMINARY RESULTS

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Keywords: cognitive impairment, screening tool, prospective cohort study

Background: Cognitive impairment and behavioural changes occur in approximately 50% of amyotrophic lateral sclerosis (ALS) patients. In 8-10% of patients, these changes are more severe, and fulfill the criteria for behavioural variant frontotemporal dementia (bv-FTD). The co-occurrence of ALS and bv-FTD results in a shorter disease duration and may interfere with decisions about life-prolonging therapies. The presence of physical disabilities and speech disturbances complicates the assessment of cognitive functions in ALS patients and may result in an overestimation of cognitive impairment. Therefore, a disease-specific cognitive screening tool is needed.

Objectives: 1. To validate a disease-specific cognitive screening tool for ALS patients, the ALS-FTD-Cog; 2. To estimate the frequency of cognitive impairment and behavioural changes in a cohort of incident ALS patients.

Methods: The ALS-FTD-Cog consists of 4 validated neuropsychological tests that cover the profile of cognitive impairment in ALS, i.e. fluency, language, verbal memory and social cognition. For validation we intend to include 110 incident ALS patients with a symptom onset of < 12 months and a forced vital capacity (FVC) of >70%, 18 ALS-FTD patients, 18 bv-FTD patients and 35 healthy volunteers. All participants undergo an extensive neuropsychological examination (executive functions, social cognition, language, verbal and visual memory, fluency and visuoperceptive functions). Two weeks later, the ALS-FTD-Cog is administered during a home visit. Measures of physical disability (ALS functional rating scale - revised (ALSFRS-R), respiratory function (FVC), depression and anxiety (hospital anxiety and depression scale (HADS)) and behavioural changes (ALS-FTD-questionnaire (ALS-FTD-Q) are administered.

Preliminary results: The ALS-FTD-Cog takes about 35 minutes to administer. Currently, 48 ALS patients, 2 ALS-FTD patients, 7 FTD patients and 35 healthy controls have participated, with a mean (SD) age of 61.3 (11.2), 57.9 (9.9), 68.7 (0.9) and 59.1 (15.1) years, respectively. The median disease duration of the ALS patients was 8 (range 4-12) months and the median ALSFRS-R score was 41 (28-47) points. The

median forced vital capacity was 89.2% (52.2-141.4). The median scores of the HADS anxiety and HADS depression were 5 (1-11) and 3.5 (1-12), respectively. The frequency of cognitive impairment, as measured with the extensive neuropsychological examination, was 42.5% in ALS patients, ie a score below the 5th percentile in 1 or more cognitive domains. 19.1% of the ALS patients had a score below the 5th percentile in 2 or more cognitive domains. The frequency of mild and severe behavioural changes was 10.9% and 10.9%, respectively, as measured with the ALS-FTD-Q.

Discussion and conclusion: Our preliminary results show that the frequency of cognitive impairment and behavioural changes in the current ALS cohort is consistent with data reported in the literature. We need to expand our cohort as planned to substantiate these data.

CW14 LONGITUDINAL VALIDATION OF THE ALS-FTD-QUESTIONNAIRE – PRELIMINARY RESULTS

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Keywords: behavioral disturbances, ALS-FTD-questionnaire, longitudinal validation

Background: Behavioral disturbances are present in approximately 30% of patients with amyotrophic lateral sclerosis (ALS). The amyotrophic lateral sclerosis - frontotemporal dementia - questionnaire (ALS-FTD-Q) is a validated screening tool to detect behavioral disturbances in ALS. For the other phenotypes within the motor neuron disease (MND) spectrum (progressive muscular atrophy (PMA) and primary lateral sclerosis (PLS)), the prevalence of behavioral disturbances, if any, is not known. Whether behavioral disturbances are progressive and whether the ALS-FTD-Q is responsive to these changes over time remains to be proven.

Objective: To determine to what extent behavioral disturbances occur in different phenotypes within the MND spectrum and to examine the responsiveness of the ALS-FTD-Q.

Methods: Patients with ALS, PMA and PLS were recruited from a specialized tertiary referral center and were assessed every 3 (ALS) to 6 (PMA and PLS) months. We examined the responsiveness by correlating the change scores of respiratory function (vital capacity, sitting and lying position, percentage predicted, and sniff nasal inspiratory pressure (SNIP), ALS functional rating scale-revised (ALSFRS-R), hospital anxiety and depression scale (HADS) and verbal fluency index (VFI) with the change scores of the ALS-FTD-Q. We used the Wilcoxon signed-rank test to determine the statistical significance of the change in scores on the ALS-FTD-Q.

Results: Thirty-two MND patients were included (21 ALS, 8 PMA, 3 PLS), one ALS patient had FTD. None of the patients developed the behavioral variant of FTD. Three and 6-months' follow up data were available from 24 and 25 patients, respectively. The ALS-FTD-Q showed longitudinal construct validity as it correlated moderately with the VFI ($r = 0.29$), and poorly with respiratory function (sitting $r = -0.25$, lying $r = -0.19$, SNIP $r = -0.07$), and ALSFRS-R ($r = -2.64$) and HADS ($r = -0.06$) at 3 and 6 months' follow up. None of the patients showed progression of behavioral disturbances over a 3-6 months' period.

Discussion and conclusions: These preliminary results provide some evidence for longitudinal construct validity of the ALS-FTD-Q and no progression of behavioral disturbances at 3 and 6 months follow-up in MND patients. We recently added the behavioral interview with a proxy (Edinburgh cognitive and behavioral ALS screen, ECAS) to our protocol in order to test the clinimetric properties of the ALS-FTD-Q against this gold standard.

CW15 LONGITUDINAL ASSESSMENT OF FRONTAL COGNITIVE IMPAIRMENT IN PATIENTS WITH MOTOR NEURON DISEASE

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Keywords: longitudinal study; cognitive impairment; frontal functions

Background: Motor neuron disease (MND) is now widely recognized as a multisystem pathology. Despite the prominence of motor symptoms, indeed, up to 50% of MND patients also manifest a broad range of neuropsychological deficits. The majority of cognitive studies of MND are cross-sectional and little is known about the longitudinal course of cognitive disturbances in these patients.

Objective: This longitudinal study tested the progression of frontal cognitive impairment in patients with MND, accounting for the effect of progressive verbal and/or physical disability.

Methods: 26 non-demented patients with recently diagnosed sporadic MND were followed prospectively with clinical and neuropsychological evaluation every 3 and 6 months respectively, for a maximum follow-up of 24 months. Cognitive assessment was performed using the MMSE, verbal fluency tests, and the Test of Attentional Performance (TAP). The TAP, which is administered through an automated computerized system, permits to investigate the whole spectrum of frontal involvement in ALS, reducing verbal and/or physical disability. Alertness (in terms of its intrinsic and phasic components), divided attention, sustained attention, behavioural control and interference tendency (in terms of stimulus-reaction incompatibility) were evaluated. Scores were analyzed in terms of performance speed and performance accuracy (valid responses and omissions). Longitudinal linear models were used to assess clinical and cognitive variable changes over time and the relationship between baseline clinical features and cognitive deterioration.

Results: During follow up, MND patients experienced a progressive worsening of motor disability, with a statistically significant decrease over time of the ALSFRSR scale score ($p<0.001$), total MRC ($p<0.001$) and ALS severity scale ($p<0.001$), and increase of the upper motor neuron score ($p<0.001$). MND patients also showed a significant deterioration of the global cognition ($p=0.04$), and several frontal measures (p ranging from <0.001 to 0.04). The TAP showed that sustained attention, behavioural control and interference tendency significantly decreased over time. The progressive cognitive decline was independent of baseline motor clinical characteristics.

Discussion and conclusions: Longitudinal analyses using computerized-based, sensitive executive measures revealed a progressive cognitive decline in MND patients, which appeared relatively early in the course of MND and is not associated with baseline motor disability. Cognitive deterioration in MND encompasses both executive performance accuracy and speed.

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CW16 PROFILING LANGUAGE IMPAIRMENT IN ALS: SPELLING

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Keywords: language, spelling, cognitive change

Background: Cognitive change may occur in up to 50% of patients with ALS. While this impairment is heterogeneous, a problem with language has recently been recognised as one of the most prevalent deficits (1). Impairment has been observed in a number of areas of language ability (2) however, recent work indicates difficulties in spelling may be a strong feature of this language impairment (3). Moreover, there is an indication that spelling errors made by patients may be observably different to those made by a non-patient population.

Objectives: To investigate language specific aspects of cognitive impairment - specifically, the suggestion of distinctive spelling errors in patients with ALS - in order to better understand their origin and to potentially utilise their profile in clinical settings.

Methods: Incorrect responses to standard spelling assessment items will serve as materials to be presented to 48 undergraduate students with no history of linguistics training. Participants will be required to use a rating scale per presented item in

order to demonstrate the degree to which the provided error appears a reasonable or understandable approximation of the correct spelling of the word. Data that has been obtained from both ALS patients with and without language impairment (determined through full neuropsychological testing, n=40), and from matched controls (n=40), will serve as materials for anonymised presentation.

Results: Data will be analysed to determine whether errors on standard spelling assessment items made by patients with ALS, compared to those of matched controls, elicit more frequent judgments indicating a 'poor approximation' from non-linguistically trained individuals. Moreover, data will be analysed with respect to type of word for which mistakes have frequently been made (4).

Discussion and conclusion: Our rigorous experimental approach to analysis of spelling difficulties of patients with ALS will complement existing investigations of changes exhibited in this ability (5,6) and contribute to the wider literature examining the emerging profile of potential language impairment in patients with ALS. We will also interpret results with respect to their influence on clinic and research practice, such as in use of identification of problems in cognition.

References:

1. Abrahams S, Newton J, Niven E, et al. Amyotroph Lateral Scler. 2014;15:9-14
2. Taylor LJ, Brown RG, Tsermentseli S, et al. J Neurol Neurosurg Psychiatry. 2013;84:494-8.
3. Niven E, Newton J, Foley J, et al. Amyotroph Lateral Scler. In press
4. Bak TH, Chandran S, Cortex. 2012;48:936-44.
5. Ichikawa H, Hieda S, Ohno H et al. European Neurology. 2010;64:148-155
6. Zago S, Poletti B, Corbo M et al. Behavioural Neurology. 2008;19(4):169-75

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CW17 SPEECH PERFORMANCE IN ALS: INFLUENCE OF LINGUISTIC CONTEXT AND CONCOMITANT LANGUAGE IMPAIRMENT

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Keywords: bulbar assessment, speech motor control, language

Background: Recent studies in healthy and disordered talkers have demonstrated that language complexity can have deleterious effects on speech motor output. One reason for this is that increasing the linguistic complexity of an utterance increases demands on the speech motor system resulting in less stable patterns of movement. An unexplored hypothesis, tested in this study, is that the linguistic complexity of spoken utterances has a similar negative impact on speech communication in persons with ALS. Further, the effect of language and speech impairments in ALS may be multiplicative in these demanding communicative contexts having far greater negative consequences on speech motor control. Bulbar assessment will be optimized by integrating the influence of linguistic factors and concomitant multisystem impairments on speech performance.

Objectives: The primary objective of this study is to identify changes to speech motor output with increasing phonological and lexical complexity of utterances in patients with ALS. A secondary objective is to determine the compounding effects of language and speech impairments on speech performance in these complex linguistic contexts.

Methods: The strategy is to recruit 10 non-demented patients with ALS and 10 age and gender-matched older controls. To test our hypothesis, we will use a speech motor control measure i.e., the spatiotemporal index (STI) to estimate changes to tongue movement patterns due to speech utterance complexity. In healthy adults, repeated productions of an utterance produce highly stable patterns of tongue movement unless communication demands increase. An electromagnetic articulograph will be used to track changes to tongue movement as a result of word complexity. For this purpose, three sensors attached to the tongue will record tongue movements as each

subject produces 32 words varying in phonological and lexical complexity. Words will vary in length, age of acquisition, phonotactic probability, neighborhood density, and sonority. Words are randomized in 12 different lists in order to compute movement variability over repeated productions of an utterance using STI (Smith and Zelaznik, 1998). Standardized language and speech tests will be administered to assess impairments to these systems.

Expected outcome: Our preliminary data from eight college-aged adults shows that tongue movement variability increases with phonological complexity and word length. We expect that patients with ALS will show significantly greater tongue movement variability than the younger and older controls for words that are lexically and phonologically more complex. These results will be discussed in light of improvements to bulbar assessment for patients with ALS.

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CW18 CLINICAL INTERVENTIONS FOR SWALLOW AND HYPERACTIVE GAG REFLEX

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Keywords: swallow, gag, respiratory

Background: ALS weakens the musculature of the upper airway affecting speech and swallow function. Symptoms include spasticity, weakness, atrophy, and hyperactive gag reflex (1). Literature review reveals very little information related to this area of symptom management.

Objectives: Assess swallow function using EAT-10. Determine interventions that affect hyperactive reflex.

Methods: Obtain EAT-10 assessment on all patients seen in ALS multidisciplinary clinic. Collect information regarding speech, swallow, respiratory function, and gag reflex sensitivity. Additional information collected to include disease onset, site of disease onset, vital signs, medications, reflex assessment, concomitant medications, respiratory function, ALSFRS-R, prescribed interventions, and patient report.

Results: Work in Progress

Discussion: Lung volume recruitment exercises have been shown to improve speech and swallow function in ALS (2). The EAT-10 Assessment Tool is a valid tool for monitoring progression of swallow function (3). Anecdotal patient reports indicate improvement in hyperactive gag reflex with Nuedexta ™. Aggressive airway clearance combined with medication management of pseudobulbar affect may be the key to maintaining swallow function and depressing the discomfort of a hyperactive gag reflex.

References:

1. Miller RG, et al. (2009). Practice parameter update: The care of the patient with amyotrophic lateral sclerosis: Drug, nutritional, and respiratory therapies (an evidence-based review). Report of the Quality Standards Subcommittee of the American Academy of Neurology. Neurology, 73(15), 1218-1226.
2. Cleary S, et al. (2007). Using lung volume recruitment in patients with ALS to improve airway clearance and increase safety while swallowing. Poster presented at the 18th International Symposium on ALS/MND, Toronto, Canada.
3. Belafsky et al. (2008). Validity and reliability of the Eating Assessment Tool (EAT-10). Ann Otol Rhinol Laryngol. 117 (12): 919-24.

CW19 THE STRAW TEST FOR ASSESSING BULBAR DYSFUNCTION IN ALS

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Keywords: bulbar assessment, bulbar deterioration, ALSFRS-R

Background: Most patients with ALS will experience bulbar symptoms throughout the disease (1). Objective assessment of bulbar dysfunction has yielded protocols for measuring progression of the disease (2, 3). The Straw Test is devised to be a convenient, easily administered, and objective bedside test for assessing oromotor dysfunction in ALS. We postulate the ability to drink using a straw as a reflection of the integrity of the bulbar functions.

Objectives: The aims of this project are (a) to provide preliminary normative values, and (b) to examine the correlation with the ALSFRS-R bulbar-function related questions.

Methods: A 7-oz Styrofoam® glass was filled with 3 and 6 ounces of water using an ounce-measuring cup for accuracy. Individuals were instructed to drink the water as fast as they could with a 24 cm long and 0.5 cm wide plastic straw. They were allowed to swallow as many times as needed. Time was taken from a starting command to the time the glass was visibly empty. All individuals were seated in a chair, arms rested on their laps, and the glass, placed over a table, so they would not have to excessively bend their heads or trunk to reach the straw. Twenty-two patients were drawn from those regularly attending to the Instituto de Ortopedia Infantil Roosevelt (Bogotá - Colombia) between February and April 2015. ALSFRS-R was obtained from all but two patients. Forty-eight non-symptomatic individuals served as the reference population. Five patients had gastrostomy and could not perform the test. The time for these patients was defined as 3 times the interquartile range. One patient that completed the 3-Oz test could not complete the 6-Oz test due to coughing and difficulties with swallowing. One patient that completed the 3-Oz test was unwilling to complete the 6-Oz test. The upper limit of normal was defined as 2SD above mean. Spearman's rho was used for assessing linear dependence between the ALSFRS-R bulbar-function related questions (speech, salivation, swallowing) and the times obtained in the 3 and 6-Oz tests.

Results: The upper limit of normal for the 3 and 6-Oz tests are 9 and 15.1 seconds, respectively. Strong linear inverse correlation was found between the ALSFRS-R bulbar-function related questions and the times obtained for both tests (Spearman's rho between -0.7 and -0.91).

Conclusions: This is a preliminary study, but the straw test may be a simple and objective tool for assessing bulbar dysfunction and progression in ALS. Further analysis of the psychometric properties of the straw test is mandatory.

References:

1. del Aguila MA, Longstreth WTJ, McGuire V et al. Neurology. 2003;60:813-819.
2. Green JR, Yunusova Y, Kuruvilla MS et al. Amyotroph Lateral Scler Frontotemporal Degener. 2013;14:494-500.
3. Yunusova Y, Green JR, Wang J et al. J Vis Exp. 2011

CW20 SPEECH GENERATING DEVICES AND YOUNG CARERS: A CASE STUDY OF TWINS CARING FOR A PARENT WITH ALS

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Keywords: young carers, speech generating device, family caregiving

Background: Approximately 80-95% of people with ALS (PALs) use some form of augmentive or alternative communication, including speech generating devices(1). For the patient, these devices provide a connection to their family and means for getting their needs met. Adult caregivers describe stress and frustration as they attempt to interact with the devices. However, it is unknown how the devices affect other family caregivers, specifically children and youth under the age of 19 who provide care to a family member ("young carers"). These isolated caregivers struggle with their parent's illness, while providing complex caregiving tasks. Dealing with a device in place of their parent's voice, may cause additional family stress and impact the psychological well-being of the young carer. Yet, no data exists describing their experience.

Objectives: This case study sought to detail the previously unknown sibling young carer experience – and how the use of a speech generating device for their parent with ALS affects the caregiving experience.

Methods: Semi-structured interviews were conducted with a set of 18-year-old twins, accessed through a Midwestern chapter of the ALS association. Qualitative data was analyzed using case explanation building techniques.

Results: Positive and negative phenomena were elicited regarding the use of a speech generating device, including: 1) voice on the machine was seen as something separate and not "actually" their mother, creating a shared sense of disobedience in the siblings, rebelling against a machine asking them to assist with her care and the home; 2) device as a catalyst for sibling support, proving an avenue to "get away" from the device, they went for drives together to cry and share; 3) served as a barrier to connection with parent. Despite providing care for their mother, they often avoided her because the device voice was irritating and simply "not her".

Discussion and conclusion: By providing the first data on how speech generating devices affect young carers, this case example describes the complex phenomena of young carers interacting with these devices in the context of caregiving. Results lay the groundwork for specific hypotheses focusing on how devices can be best configured and developed using the actual voice of the PALs to support the family, and family interactions, including those with young carers. Results further highlight the need to assess all siblings within a family affected by ALS – developing a range of supports and how to engage the young carer in technology development and use with their PALS.

References:

1. McKelvey M, Evans D, Kawai N, and Beukelman D (2012). Communication styles of Persons with ALS as recounted by surviving Partners. *Augmentative and Alternative Communication*, 2012; 28(4): 232–242

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CW21 PRESERVING LEGACY: A GUIDE TO MESSAGE BANKING IN ONE'S OWN VOICE

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Keywords: message banking, augmentative communication, assistive technology

People with motor neuron disease are superb candidates for recording messages to later incorporated into personal AAC technology, allowing personality and authentic projection of self to be expressed with a speech generating device. A model that has successfully supported people who are losing their ability to speak to preserve a part of self by using messages in their own voice is detailed. Definitions, tools and strategies will be provided.

Using one's own voice in AAC technology has been recommended for many individuals who will lose the ability to speak, yet the practice and the terms themselves are understood differently by many speech-language pathologists, physicians and potential consumers of AAC speech generating technology. This can lead patients and families to be confused or misinformed about the potential options available and may cause them to have unrealistic expectations. To add to this dilemma, the concept of voice banking is referenced on the web in confusing and conflicting ways, leading patients who face a temporary or permanent loss of speech to be confused and poorly informed. When we introduced the term 'voice banking' to the field in the early 90's, it pre-dated the existence of current technologies designed to support a person to create a synthetic voice that approximates their biological voice, a strategy that better fits the term 'voice banking'. While many people with MND are unable to participate in voice banking, they are able to MESSAGE BANK in their own voice. Indeed, many people using various technologies ranging from ipad based systems to integrated eye-tracking technology, have message banked thousands of words, phrases, sounds and stories in their own voice, intonation and emotion; preserving the legacy of self when speech production is difficult.

A link to a web-based downloadable Message Banking Guide with full details of each step, more than 50 pages of sample messages created by people with MND and details of the process will also be provided as will be links to view videos of people with MND successfully using message banking.

References:

http://america.aljazeera.com/watch/shows/techknow/articles/2014/4/10/the-silence-is-heartbreaking.html
http://america.aljazeera.com/watch/shows/techknow/articles/2014/4/9/capturing-a-lovedoneasvoice.html
https://www.youtube.com/watch?v=0xRWmxhDBQI
https://www.youtube.com/watch?v=qUcpyAQpmiM

Preserving Legacy: being Proactive with ALS, Message Banking and Low Tech

CW22 EXPLORING INDIVIDUAL QUALITY OF LIFE IN AMYOTROPHIC LATERAL SCLEROSIS (ALS)

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Keywords: individual quality of life, patients, caregivers

Background: The Schedule for the Evaluation of Individualized Quality of Life-Direct Weighting (SEIQoL-DW) assesses Individual Quality of Life (QoL) in ALS (1). The suggestion that the SEIQoL-DW measures a concept other than QoL in ALS highlights a need for further research into users' perceptions of this individual QoL measure overtime (2).

Objectives: This study aims to identify factors influencing individual QoL in patients and caregivers over time and explore the perceived reasons given for change.

Method: As part of a large longitudinal study, the SEIQoL-DW was administered to patients and caregivers at three time points over the course of 12-18 months. Respondents nominate the five areas (cues) of life most important to their QoL, rate the level of functioning of each cue on a scale of 0 (worst possible) to 100 (best possible), and then indicate the relative importance of the cues to their QoL. Ten patient-caregiver dyads were studied to provide additional information on the SEIQoL-DW during a follow-up interview. They rated the level of functioning and relative importance of the cues they had nominated at baseline, allowing for comparison of similar cue domains over time. If there was a difference of at least 20% in level of functioning or 10% in relative weight, respondents were asked to explain the reason for this. Statistical analysis was conducted using SPSS-22, with a thematic analysis of reasons for change.

Results: An overview of patient-caregiver QoL, including SEIQoL-DW index scores, cues nominated, changes in level of functioning and relative weight at follow-up compared to baseline and reasons for any change are presented. A preliminary analysis shows the most frequent cues are family, health, holidays and friends. For most respondents the nominated cues remain stable in level of functioning and relative weight over time. When there are changes it is more often in terms of level of functioning than relative weight (importance).

Discussion and conclusions: This study identifies factors influencing individual QoL in ALS. Exploring patient and caregiver reasons for change allows for an additional perspective when assessing individual quality of life using the SEIQoL-DW measure.

References:

1. Neudert C, Wasner M, and Borasio GD (2004). Individual quality of life is not correlated with health-related quality of life or physical function in patients with amyotrophic lateral sclerosis. Journal of palliative medicine, 7(4), 551-557.
2. Felgoise SH, Stewart JL, Bremer BA, et al. (2009). The SEIQoL-DW for assessing quality of life in ALS: strengths and limitations. Amyotrophic lateral sclerosis, 10(5-6), 456-462.

Acknowledgements: This study is funded by the Health Research Board (HRB) Dublin as part of the HRB Interdisciplinary Capacity Enhancement Awards.

CW23 PATIENT-CENTRED DECISION SUPPORT TOOLS FOR ALS/MND

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Keywords: patient-centred care, decision-making, multidisciplinary care

Background: ALS/MND patients and their families are continually required to make decisions for symptom management and quality of life as their condition deteriorates. Key decisions include: use of disease-slowing medication; assisted ventilation; artificial nutrition and hydration; access to palliative

care and genetic testing. ALS/MND presents many challenges to well-timed and effective decision-making. Few evidence-based treatments are available to slow or alleviate symptom progression. Moreover, in countries where physician-assisted suicide is permitted, patients may need to decide on hastening death if their condition becomes unbearable. Health professionals are unsure of the optimal timing of discussing treatment decisions with patients and families. Patients are frequently overwhelmed by the diagnosis, and may take time to come to terms with their prognosis. Symptom management information provided to patients' is frequently confronting. Currently, there are no decision support tools available to help ALS/MND patients to manage their condition. Additionally, patient-centred decision-making approaches may be novel in some cultural contexts.

Objective: Our aim is to create tools that enable ALS/MND patients to engage with health professionals, to make well-timed and well-informed decisions over the course of their disease.

Method: Two sets of decision-making tools are being developed in a collaborative project between German and Australian research groups in 2015-16. The tools will accommodate linguistic, cultural and legal considerations of ALS/MND patients and families in Germany and Australia, and will be published in both English and German. The project will use a two stage process to first develop and then validate the tools, using an expert panel of patients, family members, health professionals and researchers in each country. The tools will be developed according to international best practice standards of the International Patient Decision Aid Standards criteria. They will draw on ALS/MND evidence-based research, best practice and clinical guidelines to assist patient decisions and promote health literacy.

Expected conclusion and future directions: Content of the tools will address life prolonging and life shortening measures as appropriate to each context. In addition, a tool discussing genetic testing will be offered to patients who are possible gene carriers of familial ALS/MND. Completed tools will provide information on the benefits, risks, costs and legal issues for each option, and incorporate patients' personal values and family wishes into their decision. These decision tools will provide a useful and informative package to clarify patients' preferences, and assist patients to make difficult decisions in a timely way. Besides supporting decision making in ALS/MND, the tools, and their process of development, could have wider clinical application for a range of degenerative neurological conditions, and have potential to be expanded to countries and language groups beyond the initial collaboration.

CW24 ONLINE SHARING AND SUPPORT AMONG USERS OF A MND FORUM

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Keywords: online, sharing, web

Background: There are increasing opportunities for individuals with Motor Neurone Disease (MND) and their carers to seek information and share knowledge, experiences and emotions online. Although studies have examined the use of web forums within healthcare and some attention has been paid to information exchange and provision of advice (1), few studies have focused on the *processes* of sharing of information and experiences. The role of trust and empathy in mediating illness-based online sharing is also under-researched (2). This poster will report work in progress from a research project that is examining information posted to the MND Association UK online forum. The study is part of the 'A Space for Sharing' project which is investigating how people in extreme circumstances share information online and how acts of sharing impact on the development of trust and empathy.

Objectives: This project will investigate the processes (eg, emotional, practical, relational) involved in sharing in the MND Association forum, and will establish what the potential benefits are, if any, of developing trusting and empathic relationships in online environments for those living with MND.

Methods: The study involves qualitative analysis of information posted on the forum. 120 forum threads will be identified for inclusion in the analysis sample. In addition, approximately 12 semi-structured interviews will be conducted with people living with MND who use the forum. Interview accounts will offer insights into contextual factors that shape sharing practices. Both datasets will be analysed using thematic analysis.

Outcomes: The poster will report on initial themes emerging from the analysis. The findings will provide understanding about how trust and empathy can be

facilitated in online environments. They will also highlight factors that contribute to under-sharing, poor sharing and loss of trust between individuals interacting in online spaces. This will be of interest and practical use for charitable and health organisations (including the MND Association) seeking to explore innovative ways of using, designing and facilitating digital environments to deliver and/or enhance support services.

References:

1. Sillence E. Giving and Receiving Peer Advice in an Online Breast Cancer Support Group. Cyberpsychology, Behavior and Social Networking 2013; 16 (6):480-485
2. Nambisan P. Information seeking and social support in online health communities: impact on patients' perceived empathy. J Am Med Inform Assoc 2011; 18: 298-304.

Acknowledgements: Thanks to the MND Association for their support with this work. This project is part of the EMoTICON Network, funded through the following cross-council programmes: Partnership for Conflict, Crime and Security Research (lead: Economic and Social Research Council (ESRC)). Connected Communities (lead: Arts & Humanities Research Council (AHRC)): Digital Economy (lead: Engineering and Physical Sciences Research Council (EPSRC)) in partnership with: Defence Science and Technology Laboratory (Dstl): Centre for the Protection of National Infrastructure (CPNI).

CW25 LEVEL OF SATISFACTION WITH ASSISTIVE TECHNOLOGY DEVICES FOR PATIENTS WITH AMYOTROPHIC LATERAL SCLEROSIS

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Keywords: assistive technology, satisfaction, devices

Introduction: Progressive muscular weakness in the limbs, trunk, and bulbar muscles are routine symptoms within the course of Amyotrophic Lateral Sclerosis (ALS). As a result of muscular weakness, patients with ALS have physical impairments that affect their activities of daily living (ADL) performance. One of the clinical management recommendations for ALS patients with physical impairments includes the prescription of assistive devices to improve their function, maintain independence, and decrease fatigue (1-3). The purpose of this study is to identify what are the assistive technology devices that patients with ALS use and their level of satisfaction with it.

Methods: This is an ongoing survey study. The patients with ALS are being recruited from the ALS clinic at the MARC since May 6th 2015, at the University of Texas Health Sciences in San Antonio. Two self-report assessments that measure satisfaction are being used: Quebec User Evaluation Satisfaction with Assistive Technology- (QUEST 2.0) and the Functional Mobility Assessment (FMA).

Results: This is ongoing research- So far there are 10 participants enrolled. The majority of them have devices that assist them with mobility, such as canes, walkers and wheelchairs.

Conclusion: The results of this study will improve clinical evidence about patients' satisfaction with assistive technology equipment, and level of satisfaction related to the service provided to them.

References:

1. Sonenblum SE, Sprigle S, Harris FH, Maurer CL (2008). Characterization of Power Wheelchair Use in the Home and Community. Arch Phys Med Rehabil, 89:486-491.
2. Gruis KL, Wren PA, Huggins JE (2011). ALS Patients' Self-Reported Satisfaction with Assistive Technology. Muscle Nerve, 43(5): 643-647
3. Ward AL, Sanjak M, Duffy K, et al (2010). Power Wheelchair Prescription, Utilization, Satisfaction, and Cost for Patients With Amyotrophic Lateral Sclerosis: Preliminary Data for Evidence-Based Guidelines. Arch Phys Med Rehabil, 91: 268- 272

CW26 DIFFERENT METHODS OF WHEELCHAIR ACTIVATION- A SINGLE CASE STUDY

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Keywords: wheelchair, switches, assessment

As a result of muscular weakness, patients with ALS have physical impairments that affect their activities of daily living (ADL) performance. Some of the patients have a faster progression than the others. This clinical case illustrates the assessment process with a patient that is progressing (deteriorating) very fast. In a period of time of one year, the team re-assessed him multiple times. At each assessment the team reevaluated where the client had function and adjusted or changed the accessing method to meet the client's needs, so he could still perform his mobility independently and safely.

A total of four different types of activation methods between joysticks and switches were prescribed. The goal of this presentation is to discuss what was assessed in each visit and how the client and the team decided the best way for the client to activate the wheelchair and wheelchair features.

CW27 THE IMPLEMENTATION OF A SYSTEMATIC SCREENING REGARDING DRIVING CAPACITY IN PATIENTS WITH ALS. WORK IN PROGRESS

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Keywords: driving, multidisciplinary, quality improvement

Background: The ability to drive is often associated with independence and identity for disabled patients. The responsibility of the health care professionals is to discuss with patients the likelihood of the eventual cessation of driving and to create proactive transition plans to facilitate transportation. There is not one single test for assessing driving safety. The on-road evaluation remains the "gold standard". There are ways however, as clinicians that we can identify red flags in deciding when an on-road test is necessary. As part of a quality improvement initiative looking at implementing a consistent practice to assess driving capacity, the interdisciplinary team conducted a retrospective chart review in order to assess what information was being assessed to make the decisions re: cessation of driving or on road evaluation. The findings revealed the process was inconsistent, at times subjecting patients to numerous re-evaluations increasing the burden of time and cost on them.

Objective: The implementation of a new process to be used consistently amongst the health care team resulting in a decreased burden on the patient of repeated assessments and formal evaluations.

Method: The team proposed to improve the present process/method of evaluation. The following is a description of what will be included in the new evaluation process based on literature and health care professional experts, the following tools were suggested: 1) Develop questionnaires for both patient and caregiver (person who most often is present with patient); 2) The occupational therapist would use standard functional screening tests: Rapid-Pace Walk, Foot-Tap Test, Arm Reach, Head/Neck & Upper Body Rotation, TMA, TMB, Delayed Recall, Scan Test; 3) The respiratory therapist would perform standard pulmonary function test and apply results to the GOLD scale, would also track use of any respiratory aids (oxygen, BiPAP, etc); 4) ALS cognitive Behavioral Screen would be used to assess for cognitive functioning; 5) ALSFRS-R and Borg scale; 6) Medication list. The patient will be assessed at these following time points: baseline, every 6 months until termination or forfeit of license.

Results: The results presented will highlight data from the retrospective review, elaborate on the clinical experience of implementing this process, and the number patients that required retesting post implementation.

CW28 CERVICAL SPINE DISEASE MIMICKING BULBAR ALS

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Keywords: bulbar, cervical spinal stenosis, diagnosis

Introduction: Bulbar symptoms in patients suspected of ALS are typically attributed to disease involving the brainstem where the medullary centers involved with speech and swallowing reside. A corollary of this relationship is that a patient with bulbar symptoms undergoing diagnostic evaluation for suspected ALS requires imaging of the brain to exclude alternative disease processes and imaging of the cervical spine is not mandatory.

Objective: To call attention to the fact that cervical spine disease can cause bulbar symptoms as well as both upper and lower motor neuron signs.

Case report: A 73 year-old man was referred for a second opinion regarding the diagnosis of ALS. He had a 3 year history of bilateral leg weakness and dysphagia. His speech remained normal. Exam revealed proximal left leg weakness, diffuse hyperreflexia, extensor plantar responses, and normal sensory examination. EMG studies showed fibrillation potentials and/or enlarged motor unit potentials in multiple muscles in the cervical and lumbosacral regions.

Results: MRI of the cervical spine showed severe cervical spinal stenosis at the C3-4 level due to a disk osteophyte complex anteriorly and ligamentum hypertrophy posteriorly. There was associated T2 signal abnormality within the spinal cord at this level. Disk osteophyte complexes were also present at C4-5 and C5-6 levels. Prominent ventral osteophytes extended anteriorly from C4, C5, and C6 levels producing compression of the esophagus. Following cervical spine decompression surgery, his dysphagia resolved.

Conclusions: Evaluation of patients suspected of bulbar ALS should include cervical spine imaging as large ventral osteophytes may physically compress the esophagus and cause dysphagia.