Clinical Neurophysiology 128 (2017) 495–500

Contents lists available at ScienceDirect

Clinical Neurophysiology

journal homepage: www.elsevier.com/locate/clinph

Motor Unit Number Index (MUNIX) detects motor neuron loss in pre-symptomatic muscles in Amyotrophic Lateral Sclerosis

Christoph Neuwirth [a,*], Paul E. Barkhaus [b], Christian Burkhardt [a], José Castro [c], David Czell [d], Mamede de Carvalho [c], Sanjeev Nandedkar [e], Erik Stålberg [f], Markus Weber [a,g]

[a] Neuromuscular Diseases Unit/ALS Clinic, Kantonsspital St. Gallen, St. Gallen, Switzerland
[b] Medical College of Wisconsin, Milwaukee, WI, USA
[c] Department of Neurosciences, Hospital de Santa Maria, Instituto de Medicina Molecular, Faculty of Medicine, University of Lisbon, Lisbon, Portugal
[d] Kantonsspital Winterthur, Winterthur, Switzerland
[e] Natus Medical Inc, Middleton, WI, USA
[f] Institute of Neurosciences, Uppsala University, Department of Clinical Neurophysiology, University Hospital, Uppsala, Sweden
[g] Department of Neurology, University Hospital Basel, Basel, Switzerland

See Editorial, pages 482–483

ARTICLE INFO

Article history:
Accepted 20 November 2016
Available online 18 December 2016

Keywords:
MUNIX
Pre-symptomatic ALS
Biomarker
Multicentre
ALSFRS-R

HIGHLIGHTS

- In pre-symptomatic muscles MUNIX can detect motor unit loss.
- MUNIX is more sensitive to change compared to CMAP and ALSFRS-R.
- This makes MUNIX a biomarker candidate for disease progression.

ABSTRACT

Objective: Motor Unit Number Index (MUNIX) is a quantitative neurophysiological measure that provides an index of the number of lower motor neurons supplying a muscle. It reflects the loss of motor neurons in patients with Amyotrophic Lateral Sclerosis (ALS). However, it is unclear whether MUNIX also detects motor unit loss in strong, non-wasted muscles.
Methods: Three centres measured MUNIX in 49 ALS patients every three months in six different muscles (abductor pollicis brevis, abductor digiti minimi, biceps brachii, tibialis anterior, extensor digitorum brevis, abductor hallucis) on the less affected side. The decline of MUNIX in initially non-wasted, clinically strong muscles (manual muscle testing, MMT grade 5) was analysed before and after onset of weakness.
Results: In 49 subjects, 151 clinically strong muscles developed weakness and were included for analysis. The average monthly relative loss of MUNIX was 5.0% before and 5.6% after onset of weakness. This rate of change was significantly higher compared to ALS functional rating scale (ALSFRS-R) and compound muscle action potential (CMAP) change over 12 months prior to the onset of muscle weakness ($p = 0.024$).
Conclusion: MUNIX is an electrophysiological marker that detects lower motor neuron loss in ALS, before clinical weakness becomes apparent by manual muscle testing.
Significance: This makes MUNIX a good biomarker candidate for disease progression and possibly pharmacodynamics responds.

Abbreviations: ADM, abductor digiti minimi muscle; AH, abductor hallucis muscle; ALS, Amyotrophic Lateral Sclerosis; ALSFRS-R, revised amyotrophic lateral sclerosis functional rating scale; APB, abductor pollicis brevis muscle; BB, biceps brachii muscle; CI, Confidence Intervals; CMAP, compound muscle action potential; EDB, extensor digitorum brevis muscle; FDI, first dorsal interosseus muscle; LMN, lower motor neuron; MMT, manual muscle testing; MUNE, motor unit number estimation; MUNIX, motor unit number index; SD, standard deviation; TA, tibialis anterior muscle; UMN, upper motor neuron.

* Corresponding author at: Neuromuscular Diseases Unit/ALS Clinic, Kantonsspital St. Gallen, Rorschacherstrasse 95, CH-9007 St. Gallen, Switzerland. Fax: +41 714946389.
E-mail address: christoph.neuwirth@kssg.ch (C. Neuwirth).

http://dx.doi.org/10.1016/j.clinph.2016.11.026
1388-2457/

1. Introduction

Motor Unit Number Index (MUNIX) is a quantitative electrophysiological technique that provides an index of the number of functional lower motor neurons (LMN) supplying a muscle. Recent studies have demonstrated a good test-retest reliability in healthy subjects and ALS patients and its capability to track loss of functional LMNs over time (Ahn et al., 2010; Nandedkar et al., 2010, 2011; Neuwirth et al., 2011, 2015; Boekestein et al., 2012; Fathi et al., 2016).

MUNIX applies a mathematical model, using the area and power of the compound muscle action potential (CMAP) after supramaximal electrical stimulation of a mixed peripheral nerve and area and power of the surface electromyography (EMG) at different levels of voluntary isometric contraction. These values are used to compute the "ideal case motor unit count" to estimate the amount of functioning motor neurons. MUNIX is fast, non-invasive, and can be applied to any distal or proximal muscle in which a CMAP can be elicited by supramaximal electrical nerve stimulation. The method has been described in detail previously (Nandedkar et al., 2004, 2010). Measurements are usually performed in less than five minutes per muscle (Neuwirth et al., 2015, 2016).

Onset of ALS usually starts focally in the cervical/lumbar regions (limbs), bulbar region, or thoracic region, then spreads to contiguous regions (Ravits et al., 2007).

Consequently, measurements from several arm and leg muscles can provide information on the pattern of disease spread as opposed to measurements in only a single muscle (Neuwirth et al., 2015).

In a previous study, MUNIX measurements in 6 different muscles revealed a significant higher decline rate than the revised ALS functional rating score (ALSFRS-R) and was similar in different types of ALS onset (bulbar, arm, leg onset) (Neuwirth et al., 2015). However, it is not known whether MUNIX is able to detect LMN loss in strong, non-wasted (here denoted pre-symptomatic) muscles.

The aim of this study was to determine the rate of MUNIX decline prior and after the onset of weakness in initially clinical strong muscles (modified MRC manual muscle testing grade 5) and to compare MUNIX decline rates with CMAP amplitude decline rates and the ALSFRS-R, a well-established functional measure of disease progression (Kaufmann et al., 2007).

2. Methods

2.1. Subjects

ALS patients were recruited in specialised ALS centres in St. Gallen, Lisbon, and Milwaukee. The study protocol was approved by the local ethics committees. All subjects gave written informed consent.

ALS patients fulfilled the categories for possible, probable-laboratory supported, probable, or definite ALS according to the revised El Escorial criteria (Brooks et al., 2000). Patients were excluded, if they had other diseases that could influence cooperation or measurements (e.g. polyneuropathy, radiculopathy, peripheral nerve lesion, carpal tunnel syndrome, major stroke, frontotemporal dementia). Time from symptom onset (weakness, dysarthria, dysphagia, dyspnoea, gait impairment) to first measurement had to be less than 24 months. This criterion was added to avoid bias towards patients with slow progression or subjects with advanced stages and already numerous wasted muscles at study entry. Assessments and measurements were performed every three months ± two weeks scheduled at the regular clinic visits.

2.2. MUNIX procedure

To reduce systematic variability caused by electrode size and type, all centres used the same self-adhesive disposable surface ground and disc recording electrodes with 15 mm diameter (Ref 019-415200, Natus, Middleton, WI, USA) and arrangement of stimulation/recording electrodes (Barkhaus et al., 2006). Keypoint®-Classic-, Keypoint®.net- and Synergy®-electromyographies were used for measurements. Stimulation electrodes were not standardised across centres. The protocol for MUNIX measurements, model and computation has been reported in detail previously (Nandedkar et al., 2004, 2010; Neuwirth et al., 2010, 2011).

MUNIX was performed in the abductor pollicis brevis (APB), abductor digiti minimi (ADM), biceps brachii (BB), tibialis anterior (TA), extensor digitorum brevis (EDB), and abductor hallucis (AH) muscles.

A mandatory step in the procedure was to move the active recording electrode several times until the maximum CMAP amplitude with a clear negative take-off (first negative deflection) was obtained. The clinically less affected side was selected for measurements, which was generally the opposite side of symptom onset in order to prevent measurements in severely affected muscles and to reduce the risk of an early "floor-effect" (Neuwirth et al., 2015). In case of no detectable weakness in limb muscles, the right side was chosen.

2.3. Measures

At each visit, ALSFRS-R score was determined by the same ALS research nurse or neurologist. MUNIX raters were blinded to the results of the ALSFRS-R score and previously obtained CMAP and MUNIX measurements. Results of these longitudinal MUNIX measurements have partially been published (Neuwirth et al., 2015). This study aimed to analyse only pre-symptomatic muscles for which additional longitudinal data sets were available.

Manual muscle testing (MMT), using a modified 8-grade scale, was performed in all muscles prior to MUNIX measurements by an experienced ALS neurologist. The 8-grade scale was selected to distinguish different force levels (Table 1). Numeric values for statistical analysis for MMT4 + was 4.3 and consequently 3.7 for MMT4-.

For analysis of pre-symptomatic muscles, only clinically strong (MMT 5) and non-wasted muscles were considered for which at least one measurement before onset of weakness on follow up was available.

To evaluate change of MUNIX and CMAP in pre-symptomatic muscles, the last point in time with MMT grade 5 was arbitrarily set to "month 0" (M0). In case the rater documented slight clinical weakness in a muscle which was not detectable on the next follow-up visit with MMT grade 5, the last measurement with MMT grade 5 was set to M0. The relative change of MUNIX, ALSFRS-R, CMAP,

Table 1
Manual muscle testing (MMT) grading scale.

Grade	
5	Normal strength
4+	Inability to resist against maximal pressure
4	Ability to resist against moderate pressure
4–	Ability to resist against minimal pressure
3	Ability to move through full ROM AG
2	Ability to move with GE
1	Visible muscle contraction
0	No movement/contraction at all

ROM = range of movement; AG = against gravity; GE = gravity eliminated.