Clinical Neurophysiology 130 (2019) 315–319

Contents lists available at ScienceDirect

Clinical Neurophysiology

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

Motor Unit Number Index (MUNIX) of hand muscles is a disease biomarker for adult spinal muscular atrophy

René Günther [a,b], Christoph Neuwirth [c], Jan Christoph Koch [d], Paul Lingor [d,e], Nathalie Braun [c], Robert Untucht [a], Daniel Petzold [a], Markus Weber [c], Andreas Hermann [a,b,*]

[a] Department of Neurology, Technische Universität Dresden, Dresden, Germany
[b] German Center for Neurodegenerative Diseases (DZNE), Dresden, Germany
[c] Neuromuscular Disease Unit/ALS Clinic, Kantonspital St. Gallen, St. Gallen, Switzerland
[d] Department of Neurology, University Medical Center Göttingen, Göttingen, Germany
[e] Cluster of Excellence Nanoscale Microscopy and Molecular Physiology of the Brain (CNMPB), Germany

See Editorial, pages 305–306

ARTICLE INFO

Article history:
Accepted 11 November 2018
Available online 24 November 2018

Keywords:
Split hand
Spinal muscular atrophy
Motor unit number
Motor unit size
Amyotrophic lateral sclerosis

HIGHLIGHTS

- Hand muscle denervation was studied in adult spinal muscular atrophy (SMA) patients with MUNIX.
- Adult SMA patients had a pathophysiological remarkable denervation pattern of hand muscles, a 'reversed split hand'.
- MUNIX is a biomarker for upcoming questions in adult SMA.

ABSTRACT

Objective: There is still insufficient knowledge about natural history in adult spinal muscular atrophy, thus valid markers for treatment and disease monitoring are urgently needed.
Methods: We studied hand muscle innervation pattern of 38 adult genetically confirmed 5q spinal muscular atrophy (SMA) patients by the motor unit number index (MUNIX) method. Data were compared to healthy controls and amyotrophic lateral sclerosis (ALS) patients and systematically correlated to typical disease-relevant scores and other clinical as well as demographic characteristics.
Results: Denervation of hand muscles in adult SMA was not evenly distributed. By calculation of the MUNIX ratios, we identified a specific hand muscle wasting pattern for SMA which is different to the split hand in ALS. Furthermore, MUNIX parameters strongly correlated with established disease course parameters independent of disease stages.
Conclusion: We found a pathophysiological remarkable denervation pattern of hand muscles, a 'reversed split hand'. MUNIX of single hand muscles correlated well with disease severity and thus represents an easily available biomarker for adult SMA.
Significance: Our data show the power of the MUNIX method as a biomarker for upcoming questions in adult SMA.

Abbreviations: ADM, abductor digiti minimi muscle; ALS, amyotrophic lateral sclerosis; ALSFRSR, amyotrophic lateral sclerosis functional rating scale revised; APB, abductor pollicis brevis muscle; AUC, area under the curve; CMAP, compound muscle action potential; FDI, first dorsal interosseous muscle; HFMSE, Hammersmith functional motor scale expanded; MUNIX, motor unit number index; MUSIX, motor unit size index; ROC, receiver-operator characteristics; RULM, revised upper limb module; SIMUNIX, split hand motor unit number index; SMA, spinal muscular atrophy; *SMN1*, survival of motorneuron 1; Sum MUNIX, sum of APB MUNIX, ADM MUNIX and FDI MUNIX.

* Corresponding author at: Technische Universität Dresden, Fetscherstraße 74, 01307 Dresden, Germany.
E-mail addresses: rene.guenther@uniklinikum-dresden.de (R. Günther), Christoph.Neuwirth@kssg.ch (C. Neuwirth), jkoch@med.uni-goettingen.de (J.C. Koch), plingor@gwdg.de (P. Lingor), Nathalie.Braun@kssg.ch (N. Braun), Robert.Untucht@uniklinikum-dresden.de (R. Untucht), petzold.daniel89@web.de (D. Petzold), markus.weber@kssg.ch (M. Weber), andreas.hermann@uniklinikum-dresden.de (A. Hermann).

https://doi.org/10.1016/j.clinph.2018.11.009
1388-2457/

1. Introduction

5q spinal muscular atrophy (SMA) is a monogenetic, autosomal-recessive, lower motoneuron disease caused by deletion or mutation of *SMN1* (survival of motoneuron 1) which results in reduced expression of full-length SMN protein. Although no systematic data exist on treatment effects in adult SMA patients, the antisense-oligonucleotide Nusinersen was recently approved by the FDA and EMA as a disease modifying drug for SMA patients, including adults (Mercuri et al., 2018). As only limited data are available on adult SMA and because adult SMA include clinical stages from slightly to severely affected, reliable determination of disease progression, definition of treatment goals and establishment of suitable disease (progression) biomarkers are needed for a comprehensive high quality health care.

Because hand muscle function is relatively spared, the pathophysiological involvement of the SMA hand is of great interest for clinicians and patients especially in advanced disease states. In children, several electrophysiological quantifications were published: Motor Unit Number Estimation (MUNE) and compound muscle action potential (CMAP) records were used (Kaufmann et al., 2012, Gawel et al., 2015, Kolb et al., 2016). To the best of our knowledge motor unit number index (MUNIX) was not investigated in adult SMA patients so far. MUNIX is a non-invasive, electrophysiological method combining neurography and surface electromyography to estimate the remaining number of motor units. This method has a good reliability and validity to monitor loss of motor units (Neuwirth et al., 2010, Neuwirth et al., 2011, Neuwirth et al., 2018).

The aim of the current study was to examine the functional motor units of the SMA hand muscles and to explore this method as a tool to monitor disease progression in adult SMA.

2. Methods

2.1. Participants and assessments

We analyzed data from genetically confirmed, yet untreated patients with SMA type 2 (SMAII) and SMA type 3 (SMAIII), patients with sporadic amyotrophic lateral sclerosis (ALS) according to the revised El Escorial criteria and healthy controls (for details see Table 1). Patients with clinical signs of carpal tunnel syndrome, ulnar nerve entrapment or polyneuropathy were not included. Data were collected from three different centers with expertise in motoneuron diseases: Dresden (Germany), Göttingen (Germany), St.Gallen (Switzerland). For MUNIX and motor unit size index (MUSIX) generation we used the same methodology as previously described (Nandedkar et al., 2018). All raters were extensively trained by C.N. and M.W. All patients gave their informed consent and study approval was obtained by the local ethics committees (EK393122012; 10/2/17; EKSG 09/108/1B).

Table 1
Demographic and clinical characteristics of study populations. Amyotrophic lateral sclerosis functional rating scale revised (ALSFRS-R), revised upper limb module (RULM), Hammersmith functional motor scale extended (HFSME).

	Control	ALS	SMA
Total patient number	24	65	38
Datasets of APB/ADM/FDI	24/24/24	59/65/22	38/36/36
Ratio of females in %	58.3	41.5	44.7
Age in years	39.4 ± 11.7	61.8 ± 10.8	34.9 ± 11.6
Subtype at onset (n)	-	bulbar(23) spinal (42)	SMA II (11) SMA III (27)
SMN2 copy number (n)	-	-	2 copies (1) 3 copies (13) 4 copies (11) Unknown (13)
ALSFRS-R (range)	-	36 (14–48)	33 (20–44)
HFMSE (range)	-	-	16 (0–41)
RULM (range)	-	-	23 (6–37)

2.2. Statistical analysis

As the samples were not normally distributed by calculation with Shapiro–Wilk test, statistical comparisons of data between groups were made using the non-parametric Kruskal–Wallis-ANOVA followed by a post-hoc test with Bonferroni adjustment. Spearman rank correlation coefficients were used to examine correlations with a correlation coefficient of $R < 0.3$ considered as a weak, $R = 0.3–0.59$ a moderate, and $R \geq 0.6$ a strong correlation. Data were analyzed using the software programs Statistica 13.2 [StatSoft (Europe) GmbH, Hamburg, Germany] and for calculation of receiver operating characteristics (ROC) the software program SPSS statistics 25 (SPSS Inc., Chicago, IL, USA). If not mentioned otherwise, all data are displayed as means ± SD. Significance level was set at $p < 0.05$.

3. Results

3.1. Demographic and clinical characteristics

The MUNIX parameters CMAP, MUNIX and motor unit size index (MUSIX) of the right hand of 24 controls, 65 ALS and 38 SMA patients were analyzed and compared. Demographic and clinical characteristics of the study populations are shown in Table 1. SMA and controls do not differ regarding sex ($p = 0.37$) and age ($p = 0.24$).

3.2. Characteristics of hand muscle denervation in adult SMA

Abductor pollicis brevis muscle (APB): CMAP and MUNIX were significantly reduced in SMA (CMAP: 8.1 ± 2.9 mV, $p < 0.05$; MUNIX: 103.1 ± 63.1, $p < 0.001$) compared to controls (CMAP: 10.5 ± 2.3 mV; MUNIX: 184.8 ± 52.3). However, CMAP and MUNIX of SMAIII (CMAP: 9.3 ± 2.2 mV, $p = 1.0$; MUNIX: 132.1 ± 50.9, $p = 0.18$) did not significantly differ to controls while SMAII (CMAP: 5.2 ± 2.3 mV, $p < 0.001$; MUNIX: 31.6 ± 14.4, $p < 0.0001$) had severely reduced values. MUSIX was significantly increased in SMA (111.8 ± 79.7, $p < 0.0001$) compared to controls (58.1 ± 8.7).

Abductor digiti minimi muscle (ADM): CMAPs and MUNIX values were strongly decreased in SMA (CMAP: 5.2 ± 4.0 mV, $p < 0.0001$; MUNIX: 49.2 ± 44.1, $p < 0.0001$), including both SMAIII (CMAP: 6.7 ± 3.7, $p < 0.0001$; MUNIX: 61.3 ± 44.6, $p < 0.0001$) and SMAII (CMAP: 1.3 ± 0.7, $p < 0.0001$; MUNIX: 12.9 ± 7.0, $p < 0.0001$), compared to controls (CMAP: 11.8 ± 1.9; MUNIX: 183.3 ± 40.6). In two patients of SMAII denervation process was advanced to a degree that we could not detect a sufficient CMAP of the ADM. MUSIX of ADM was significantly higher in SMA (116.5 ± 44.8, $p < 0.0001$) compared to controls (65.8 ± 10.8).

First dorsal interosseous muscle (FDI): FDI was strongly affected in SMA (CMAP: 6.0 ± 5.0; MUNIX: 65.7 ± 56.0, $p < 0.0001$), including both SMAIII (CMAP: 7.8 ± 5.0, $p < 0.0001$; MUNIX: 85.4 ± 56.4, $p < 0.0001$) and SMAII (CMAP: 1.9 ± 0.9, $p < 0.0001$; MUNIX: 21.1 ± 13.5, $p < 0.0001$), compared to controls (CMAP: 18.2 ± 3.7; MUNIX: 293.0 ± 61.8). FDI MUSIX was significant higher in SMA (97.5 ± 35.8, $p < 0.0001$) compared to controls (63.3 ± 12.6). For summary see Fig. 1.

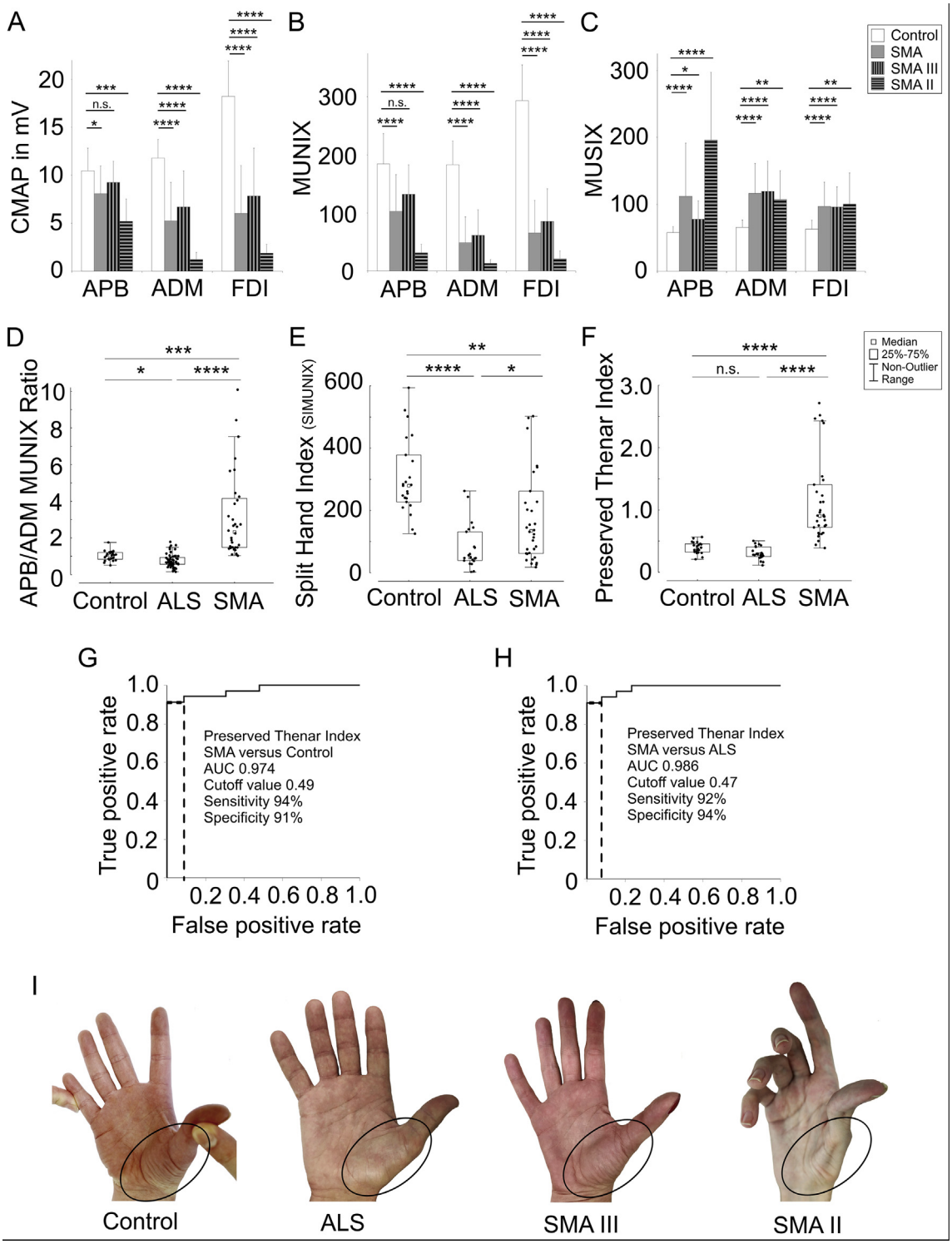

Fig. 1. Characteristics of hand muscle denervation and the reverse split hand in SMA. Depicted are histograms of CMAP (A), MUNIX (B) and MUSIX (C) of APB, ADM, FDI of controls, all SMA (SMA), and subgroups of only SMA type 3 (SMAIII) and only SMA type 2 (SMAII). The ratio of APB/ADM MUNIX shows the relation between ADM and APB MUNIX and indicate the split hand pattern in ALS and a reversed split hand pattern in SMA (D). The 'Split Hand Index (SIMUNIX)' ((APB MUNIX × FDI MUNIX)/ADM MUNIX) separate ALS from controls, but not from SMA (E). The 'Preserved Thenar Index' (APB MUNIX/(ADM MUNIX + FDI MUNIX)) differentiate SMA patients from controls and ALS (F). (D),(E),(F) are depicted as box plots with raw data (one dot represents one patient). Receiver-operator characteristic (ROC) curves for the diagnosis of SMA versus controls (G) and versus ALS (H). Representative examples of a healthy control with normal hand pattern and intact function, the ALS palm with thenar atrophy and loss of abductor function as well as the SMA palm with relatively preserved thenar muscle volume and preserved abductor function (I). $^{****}p < 0.0001$, $^{***}p < 0.001$, $^{**}p < 0.01$, $^{*}p < 0.05$, not significant (n.s.).

3.3. Selective vulnerability of hand muscles in adult SMA – the "reverse split hand"

The MUNIX split hand index (SIMUNIX = (APB * FDI)/ADM) was established to be a diagnostic tool to discriminate ALS from healthy controls as well as disease mimics and quantifies the pronounced loss of the APB and FDI motor units in relation to the relatively well preserved ADM (Menon et al., 2013, Kim et al. , 2016). Motor unit loss in hand muscles in adult SMA was not evenly distributed and seemed to be reversed compared to the pattern in ALS. While in controls, APB and ADM had the same amount of motor units (1.0 ± 0.3), this ratio was significantly different in SMA (3.9 ± 5.5) and ALS (0.8 ± 0.4) displaying a diametrical pattern ($p < 0.0001$) (Fig. 1D). By calculating the split hand index (SIMUNIX), the ALS group (82.9 ± 70.4) could be separated very well from controls (305.3 ± 119.3, $p < 0.0001$), but has only a weak difference to SMA (253.6 ± 433.2, $p < 0.05$) (Fig. 1E). By calculating the 'Preserved Thenar Index' (APB MUNIX/(FDI MUNIX + ADM MUNIX)), SMA (1.2 ± 0.8) showed a highly significant difference to ALS (0.3 ± 0.1, $p < 0.0001$) and controls (0.4 ± 0.1, $p < 0.0001$). A cutoff value of 0.49 differentiated SMA from controls as well as ALS with a sensitivity and specificity over 90% (Fig. 1F–H). The 'Preserved Thenar Index' calculated with CMAP values (APB CMAP/(FDI CMAP + ADM CMAP)) showed a lower performance in diagnostic accuracy for discrimination of the groups, the area under the curve (AUC) for CMAP-'Preserved Thenar Index' (SMA versus controls: AUC 0.91; SMA versus ALS: AUC 0.95) was lower in comparison to AUC for MUNIX-'Preserved Thenar Index' (SMA versus controls: AUC 0.97; SMA versus ALS: AUC 0.99).

3.4. MUNIX reflect disease severity in patients suffering from SMA

As there is no consensus about a valid score of severity in adult SMA, we surveyed the well known amyotrophic lateral sclerosis functional rating scale revised (ALSFRS-R), the SMA specific tests Hammersmith functional motor scale expanded (HFMSE) and revised upper limb module (RULM), hand muscle strength with a hand dynamometer and forced vital capacity (FVC) with a spirometer in a subgroup of SMA patients. Spearman rank correlation coefficients between the scores, clinical parameters, demographic parameters and the MUNIX parameters are shown in Table 2. MUNIX of APB and FDI showed strong, MUNIX of ADM moderate correlations to the disease scores (ALSFRS-R, HFMSE, RULM) and hand muscle strength. FVC correlated strongly to FDI MUNIX and moderate to APB and ADM MUNIX. Age only moderately correlated to ADM MUNIX while no correlations to FDI MUNIX or APB MUNIX were found. BMI did not correlate to MUNIX parameters. To include all MUNIX data of APB, ADM and FDI in one value, we calculated the sum (Sum MUNIX). Sum MUNIX strongly correlated to ALSFRS-R, HFMSE, RULM and hand muscle strength and moderately to FVC. The Preserved Thenar Index did not show any correlations to the disease scores, clinical parameter and demographic parameters. APB MUNIX and APB CMAP ($N = 38$, $R = 0.82$, $p < 0.0001$), ADM MUNIX and ADM CMAP ($N = 36$, $R = 0.94$, $p < 0.0001$) as well as FDI MUNIX and FDI CMAP ($N = 36$, $R = 0.94$, $p < 0.0001$) correlated strongly with each other.

4. Discussion

Our study has two remarkable results. First, adult SMA patients suffer from a 'reversed split hand' phenomenon, which discriminates SMA from ALS and controls with a high sensitivity and specifity. Interestingly, this phenomenon was also seen in clinically less affected hand muscles and did not correlate with disease progression rendering it an ideal diagnostic tool. Second, MUNIX of single hand muscles correlated well with disease severity and thus represents an easily available biomarker for adult SMA, still valid when

Table 2
Correlations of CMAP, MUNIX and MUSIX data to clinical and demographic characteristics of SMA. Spearman rank correlation coefficients with $R < 0.3$ considered as a weak, $R = 0.3–0.59$ a moderate, and $R \geq 0.6$ a strong correlation. Body mass index (BMI), forced vital capacity (FVC), amyotrophic lateral sclerosis functional rating scale revised (ALSFRS-R), revised upper limb module (RULM), Hammersmith functional motor scale expanded (HFSME). Number of patients (N), $^{****}p < 0.0001$, $^{***}p < 0.001$, $^{**}p < 0.01$, $^{*}p < 0.05$, not significant (n.s.).

	APB CMAP	ADM CMAP	FDI CMAP	APB MUNIX	ADM MUNIX	FDI MUNIX	Sum MUNIX	APB MUSIX	ADM MUSIX	FDI MUSIX	Preserved Thenar Index
Age	N=38; n.s.	0.34 N=36; P<0.05	N=36; n.s.	N=38; n.s.	0.39 N=36; P<0.05	N=36; n.s.	N=34; n.s.	N=38; n.s.	N=36; n.s.	N=36; n.s.	N=34; n.s.
BMI	N=20; n.s.	N=20; n.s.	N=20; n.s.	N=20; n.s.	N=20; n.s.	N=20; n.s.	N=20; n.s.	N=20; n.s.	N=20; n.s.	N=20; n.s.	N=20; n.s.
FVC	N=26; n.s.	0.67; N=26; p<0.001	0.60; N=26; p<0.01	0.46; N=26; p<0.05	0.58; N=26; p<0.01	0.65; N=26; p<0.001	0.59 N=26 p<0.01	-0.42; N=26; p<0.05	N=26; n.s.	N=26; n.s.	N=26; n.s.
ALSFRS-R	0.62; N=31; p<0.001	0.69; N=30; p<0.0001	0.67; N=31; p<0.0001	0.77; N=31; p<0.0001	0.57; N=29; p<0.01	0.65; N=31; p<0.0001	0.70 N=29; p<0.0001	-0.75; N=31; p<0.0001	N=29; n.s.	N=31; n.s.	N=29; n.s.
HFMSE	0.61; N=20; p<0.01	0.84; N=20; p<0.0001	0.81; N=20; p<0.0001	0.80; N=20; p<0.0001	0.57; N=20; p<0.01	0.86; N=20; p<0.0001	0.83; N=20; p<0.0001	-0.58; N=20; p<0.01	N=20; n.s.	N=20; n.s.	N=20; n.s.
RULM	0.52; N=20; p<0.05	0.71; N=20; p<0.001	0.73; N=16; p<0.001	0.77; N=20; p<0.0001	N=20; n.s.	0.81; N=20; p<0.0001	0.77; N=20; p<0.0001	-0.64; N=20; p<0.01	N=20; n.s.	N=20; n.s.	N=20; n.s.
Hand muscle strength	0.66; N=18; p<0.01	0.78; N=18; p<0.001	0.84; N=18; p<0.0001	0.87; N=18; p<0.0001	0.56; N=18; p<0.05	0.90; N=18; p<0.0001	0.88; N=18; p<0.0001	-0.74; N=18; p<0.001	N=18; n.s.	N=18; n.s.	N=18; n.s.

classical SMA scores loose its feasibility because of severity of symptoms.

Motor unit loss of hand muscles is present in adult SMA patients from slightly to severely affected individuals. The most affected muscle in SMA was the FDI, followed by the ADM. Interestingly the APB was relatively well preserved. For ALS a typical 'split hand' phenomenon is well known and comprises the weakness of the thenar/FDI group while the hypothenar group is relatively spared. It is speculated that this phenomenon is due to the cortical representation of ALS pathology (Weber et al., 2000, Eisen et al., 2017). In contrast, we observed a reversed pattern in adult SMA patients, a 'reverse split hand'. We showed that FDI and ADM were much more impaired than the APB and that the 'Preserved Thenar Index' can discriminate the pattern of SMA from ALS and controls with a high sensitivity and specifity. Whether this index is unique for SMA or rather a general pattern of lower motoneuron diseases has to be further evaluated. All of the measured hand muscles are innervated from the same cervical spinal cord segments (C7-TH1), but the controlling motoneuron somas are organized in different motoneuron pools. These motoneuron pools are maybe disease selective vulnerable, e.g. due to the developmental patterning of morphogenetic cues (Maden, 2006).

A better diagnostic accuracy for MUNIX in comparison to CMAP concerning calculation of the Split hand index in ALS was already shown by Kim et al. (Kim et al., 2016). The diagnostic accuracy of the 'Preserved thenar index' calculated by MUNIX revealed a better performance in comparison to CMAP in our study also, supporting its methodical power.

MUNIX of the hand muscles strongly correlated with validated disease scores. Therefore, MUNIX values of the hand muscles reflect well the overall disability and state of disease progression in adult SMA. The phenotypical variety in adult SMA range from mild with moderate disabilities in daily life to very severe with the need of a 24 hour assistant support. This means, that disease progression and or treatment effects in severely affected patients can – if at all – only be monitored by RULM and not with HFMSE or 6-minute walk test. Furthermore, mild affected patients can score full points in these scores disabling their use as progression biomarker in those. This phenotypic diversity makes it difficult to monitor disease progression in adult SMA patients. One clinical phenomenon in SMA is the vulnerability of proximal muscles and the clinically relatively preserved hand function. However, we show that motor unit loss is already present in clinically unaffected hand muscles and motor unit numbers of hand muscles can still be recorded also in very progressed disease stages. Monitoring motor units of hand muscles with MUNIX might therefore be a promising disease progression marker allowing to include many adult patients. The higher sensitivity of MUNIX in comparison to other established disease scores like ALSFRS-R and manual muscle testing was already shown for patients with ALS (Neuwirth et al., 2017), additionally underpinning its strength. Finally, in contrast to most of the currently recommanded disease scores, MUNIX is a patient unbiased method.

Longitudinal studies are needed to assess whether measuring one muscle – and if which one – or which of the measures presented turn out to be the best biomarker. Furthermore, systematic comparison of different techniques estimating motor unit numbers (e.g. MUNE versus MUNIX) in adult SMA are needed.

Taken together, reading the patient's palm with MUNIX might be an unbiased, valid tool for early differential diagnosis and disease progression monitoring in adult SMA patients. This easy-to-use, well-tolerated method may help to better understand natural history and increase the quality in patient-individualized medical guidance of adult SMA patients and give new insights of motoneuron disease pathophysiology.

Author contributions

R.G. and A.H. were responsible for study concept and design. All authors were responsible for acquisition and analysis of data. R.G. and A.H. were responsible for drafting a significant proportion of the manuscript and figures.

Declaration of interest

Authors declare no conflict of interests.

Acknowledgements

We thank the patients and their families for contributing to this study.

References

Eisen A, Braak H, Tredici KD, Lemon R, Ludolph AC, Kiernan MC. Cortical influences drive amyotrophic lateral sclerosis. J Neurol Neurosurg Psychiatry 2017;88:917–24.

Gawel M, Kostera-Pruszczyk A, Lusakowska A, Jedrzejowska M, Ryniewicz B, Lipowska M, et al. Motor unit loss estimation by the multipoint incremental MUNE method in children with spinal muscular atrophy–a preliminary study. Neuromuscul Disord 2015;25:216–21.

Kaufmann P, McDermott MP, Darras BT, Finkel RS, Sproule DM, Kang PB, et al. Prospective cohort study of spinal muscular atrophy types 2 and 3. Neurology 2012;79:1889–97.

Kim DG, Hong YH, Shin JY, Park KH, Sohn SY, Lee KW, et al. Split-hand phenomenon in amyotrophic lateral sclerosis: a motor unit number index study. Muscle Nerve 2016;53:885–8.

Kolb SJ, Coffey CS, Yankey JW, Krosschell K, Arnold WD, Rutkove SB, et al. Baseline results of the NeuroNEXT spinal muscular atrophy infant biomarker study. Ann Clin Transl Neurol 2016;3:132–45.

Maden M. Retinoids and spinal cord development. J Neurobiol 2006;66:726–38.

Menon P, Kiernan MC, Yiannikas C, Stroud J, Vucic S. Split-hand index for the diagnosis of amyotrophic lateral sclerosis. Clin Neurophysiol 2013;124:410–6.

Mercuri E, Darras BT, Chiriboga CA, Day JW, Campbell C, Connolly AM, et al. Nusinersen versus sham control in later-onset spinal muscular atrophy. N Engl J Med 2018;378:625–35.

Nandedkar SD, Barkhaus PE, Stalberg EV, Neuwirth C, Weber M. Motor unit number index (MUNIX): guidelines for recording signals and their analysis. Muscle Nerve 2018;58:374–80.

Neuwirth C, Barkhaus PE, Burkhardt C, Castro J, Czell D, de Carvalho M, et al. Motor Unit Number Index (MUNIX) detects motor neuron loss in pre-symptomatic muscles in Amyotrophic Lateral Sclerosis. Clin Neurophysiol 2017;128:495–500.

Neuwirth C, Braun N, Claeys KG, Bucelli R, Fournier C, Bromberg M, et al. Implementing Motor Unit Number Index (MUNIX) in a large clinical trial: real world experience from 27 centres. Clin Neurophysiol 2018;129:1756–62.

Neuwirth C, Nandedkar S, Stalberg E, Barkhaus PE, Carvalho M, Furtula J, et al. Motor Unit Number Index (MUNIX): a novel neurophysiological marker for neuromuscular disorders; test-retest reliability in healthy volunteers. Clin Neurophysiol 2011;122:1867–72.

Neuwirth C, Nandedkar S, Stalberg E, Weber M. Motor unit number index (MUNIX): a novel neurophysiological technique to follow disease progression in amyotrophic lateral sclerosis. Muscle Nerve 2010;42:379–84.

Weber M, Eisen A, Stewart H, Hirota N. The split hand in ALS has a cortical basis. J Neurol Sci 2000;180:66–70.