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ORIGINAL COMMUNICATION

Nerve ultrasound in the differentiation of multifocal motor neuropathy (MMN) and amyotrophic lateral sclerosis with predominant lower motor neuron disease (ALS/LMND)

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Abstract The objective of the study was to investigate nerve ultrasound (US) in comparison to nerve conduction studies (NCS) for differential diagnosis of amyotrophic lateral sclerosis with predominant lower motoneuron disease (ALS/LMND) and multifocal motor neuropathy (MMN). A single-center, prospective, examiner-blinded cross-sectional diagnostic study in two cohorts was carried out. Cohort I: convenience sample of subjects diagnosed with ALS/LMND or MMN (minimal diagnostic criteria: possible ALS (revised EL-Escorial criteria), possible MMN (European Federation of Neurosciences guidelines). Cohort II: consecutive subjects with suspected diagnosis of either ALS/LMND or MMN. Diagnostic US and NCS models were developed based on ROC analysis of 28 different US and 32 different NCS values measured in cohort I. Main outcome criterion was sensitivity/specificity of these models between ALS/LMND and MMN in cohort II. Cohort I consisted of 16 patients with ALS/LMND and 8 patients with MMN. For cohort II, 30 patients were recruited, 8 with ALS/LMND, 5 with MMN, and 17 with other diseases. In cohort I, the three best US measures showed higher mean ± SD areas under the curve than the respective NCS measures (0.99 ± 0.01 vs. 0.79 ± 0.03, $p < 0.001$; two-sided t test). The US model with highest measurement efficacy (8 values) and diagnostic quality reached 100 % sensitivity and 92 % specificity for MMN in cohort II, while the respective NCS model (6 values, including presence of conduction blocks) reached 100 and 52 %. Nerve US is of high diagnostic accuracy for differential diagnosis of ALS/LMND and MMN. It might be superior to NCS in the diagnosis of MMN in hospital-admitted patients with this differential diagnosis.

Keywords Nerve ultrasound · ALS · MMN · Diagnosis · Nerve conduction studies

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Introduction

Differential diagnosis of amyotrophic lateral sclerosis with leading lower motoneuron disease (ALS/LMND) and multifocal motor neuropathy (MMN) can be complicated, particularly in early disease stages [1, 2]. Although upper motoneuron (UMN) involvement makes ALS in many cases distinguishable from MMN, clinical presentation of ALS can be anywhere on a continuum from pure UMN to pure LMN involvement [3, 4]. At the same time, in MMN the hallmark finding of pure motor conduction blocks in

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nerve conduction studies (NCS) can be missing in up to 40 % of patients [5, 6]. Correct differential diagnosis of these two diseases is of high therapeutic and prognostic importance, since ALS represents an incurable disease with a median survival of less than 2 years [7], but MMN responds to immunomodulatory treatment and has a good long-term prognosis [8].

Nerve ultrasound (US) is a rapidly evolving technique in the diagnosis of peripheral nerve lesions such as carpal tunnel syndrome (CTS) [9]. Although US is not yet established as a routine clinical application in the diagnosis of polyneuropathies, several studies have shown that particularly demyelinating neuropathies are characterized by prominent morphologic changes, providing a basis for the differentiation from axonal neuropathies or healthy controls [10, 11]. MMN has a complex, possibly antibody-mediated pathology involving axonal degeneration concentrated at the nodes of Ranvier, demyelination and inflammatory infiltrates in some cases [12, 13]. In US, this corresponds to an increase in US-measured nerve cross-sectional area (CSA) [10], whereas ALS shows a decrease in CSA, possibly reflecting axonal degeneration [14].

The aim of this prospective, monocenter, rater-blinded diagnostic study was to develop a diagnostic US protocol for the differentiation of ALS/LMND and MMN and to compare the performance of this US protocol with a standard NCS protocol. The primary hypothesis was that US will be superior to NCS in this diagnostic question. One recently published study supports this hypothesis, but has chosen a different testing approach and has not compared NCS [15].

Subjects and methods

Subjects and group assignment

This single-center, prospective, examiner-blinded cross-sectional diagnostic study was performed at the Department of Neurology, Technische Universität Dresden, Saxony, Germany. Two study cohorts were recruited according to the following criteria (see Online Resource Fig. 1 and 2 for study flow diagrams).

Cohort I Cohort I was recruited as a convenience sample at our specialized outpatient clinic for patients with ALS and inflammatory neuropathies between August 2013 and October 2014. Inclusion criteria were the diagnosis of ALS/LMND or MMN. For ALS, minimal diagnostic criteria to be met were those of possible ALS according to revised El-Escorial criteria [16]. Predominant LMN involvement was defined as presence of paresis without clinical or electrophysiological sings of upper motor neuron (UMN) involvement, such as increased muscle tone, spasticity, presence of pathological reflexes or increased central motor latency in magnetic-evoked potentials; preservation of muscle tendon reflexes in paretic regions was not considered a sign of predominant UMN involvement. For MMN, minimal diagnostic criteria were those of possible MMN according to the European Federation of Neurosciences/Peripheral Nerve Society (EFNS/PNS) guidelines [8].

Cohort II Cohort II is a consecutive sample of all patients admitted between August 2013 and October 2014 with a suspected diagnosis of ALS/LMND or MMN. For study eligibility, this differential diagnostic focus had to be reconfirmed by the attending senior physician during initial clinical ward rounds.

Patients being not able to follow the study protocol due to advanced impairment (e.g., due to severe dyspnea) were excluded. All patients provided written informed consent, and the study was approved by the institutional review board.

Procedures

Diagnostic criteria for cohort I were verified by chart review and by consultation of the attendant senior neurologist in charge. Basic demographic data were recorded (age, sex, disease duration and time since diagnosis in months). For clinical signs and symptoms, the following scales were employed: Medical Research Council Sum Score (MRC-SS, all participants) [17], Modified Rankin Scale (mRS, all participants) [18], revised ALS functional rating scale (r-ALSFR, groups 1 and 3) [19], and overall disability sum score (ODSS, group 2 and 3) [18]. Grip force was documented with a dynamometer (Jamar hydraulic handgrip dynamometer, Lafayette Instrument Inc., Loughborough, UK).

Motor and sensory NCS of median, ulnar, radial, tibial, peroneal and sural nerves and f-responses of the median, ulnar and tibial nerve were obtained on a Keypoint, Software Version 3.5 (Medtronic, Meerbusch, Germany) by experienced examiners (S.F., M.W.) blinded to group assignment. In addition to the measurement values, the presence of motor conduction blocks [defined as a reduction of >50 % in amplitude or area under the curve (AUC)] was determined and included in the diagnostic NCS models (see below). Patients were examined unilaterally on the clinically more affected side.

Both transverse and longitudinal US scans were obtained in 11 nerves at 13 different sites to record CSA and diameter. Three images were taken at each measurement site, and the mean CSA and diameter were used. Measurement sites were chosen according to a highly standardized and reproducible study protocol proposed by Zaidman and co-workers [20]. It consists of measurements

distant from entrapment sites, determined by osseous reference points. Median and ulnar nerve: ¾ of the distance between the medial epicondyle of the humerus to the ulnar styloid process with the elbow orthogonally flexed; median, radial and ulnar nerve: two-thirds of the lateral tip of the acromion to the lateral epicondyle of the humerus; tibial and sural nerve: three-fourths of the distance from the head of the fibula to the lateral malleolus; sciatic nerve: two-thirds from the greater trochanter to the lateral epicondyle of the femur; cervical nerve roots 5–7: interscalenic gap; superior trunk: supraclavicular fossa; vagal nerve: halfway between carotid bifurcation and clavicle. US machines used were: Aplio MX, linear transducer, 8 to 18 MHz (Toshiba, Neuss, Germany) and MyLab Five, linear transducer, 10 to 18 MHz (Esaote Biomedica, Cologne, Germany). Frequency was varied between 10 and 18 MHz to obtain best resolution depending on the depth of the nerve of interest. Examinations were performed by two experienced investigators (J.L. K.F.L.), blinded to group assignment.

Statistical analysis

Statistical significance was defined by $p < 0.05$ for all tests. If not otherwise stated, numbers given are mean and standard deviation (SD), and the p value derived from a two-sided t test. Normal distribution of continuous data was tested with Kolmogorov–Smirnov test. If data were normally distributed, two-sided t tests were used for all two-group comparisons, otherwise a univariate ANOVA with a post hoc two-sided t test. If data were not normally distributed or ordinal, a Mann–Whitney U test or a Kruskal–Wallis test with a post hoc Mann–Whitney U test was used, respectively. All post hoc tests were Bonferroni adjusted for α-inflation. For categorical data, Fisher's exact test was used, in case of three groups with a Freeman–Halton extension. Generalized linear models (GLM) were used to assess possible confounding effects of unbalanced baseline characteristics. For calculation of mean z scores of values from different body regions or nerve subtypes in cohort I, original data were z-transformed. Correlations between mean z scores of US and NCS values and clinical scales were assessed by Pearson's correlation coefficient (a coefficient $r < 0.3$ was considered a weak, $r = 0.3$–0.59 a moderate, and $r \geq 0.6$ a strong correlation). Receiver operating curve (ROC) analysis was performed to determine the area under the curve (AUC) with 95 % confidence intervals (95 % CI) for single untransformed measurements and mean z scores.

The following selection criteria were used for development of diagnostic models to be tested in cohort II. Untransformed US or NCS measurement values were chosen based on highest AUC in ROC analysis in cohort I. For US, measurement values were either included if their AUC was ≥0.9 or the lower 95 % confidence interval was >0.5. For NCS, inclusion criteria had to be more liberal because of the much lower AUC of single measurement values; values were included if AUC was ≥0.6 or the lower 95 % confidence interval was >0.5. Cutoff values were chosen from the respective ROC analysis in cohort I at Youden index, at ≥80 or at ≥90 % specificity. Based on these cutoff values, the number of pathological measurements of each patient in cohort II was determined and counted. For NCS models, the presence of conduction blocks was added. The total number of pathological values required was chosen to provide 100 % sensitivity for MMN in cohort II. All analyses were performed with SPSS Version 22 (IBM, Ehningen, Germany).

Results

Cohort I consisted of 24 subjects including 16 patients with ALS and 8 with MMN (for demographics, see Table 1 and Online Resource Figures 1, 2 for study flow diagram). 5 of the ALS patients in cohort I (31 %) had a clinically definite diagnosis, 4 (25 %) a clinically probable, 3 (19 %) a clinically probable-laboratory supported, and 4 (25 %) a clinically possible according to revised El-Escorial criteria [16]. Of the MMN patients, 6 (75 %) had definite MMN and 2 (25 %) possible MMN according to EFNS/PNS guidelines.

During the recruitment period for cohort II, 768 consecutive patients were admitted to the general neurology ward of the study center. 31 patients (4 %) had a suspected diagnosis of either ALS or MMN after initial attending senior physician consultation and thus were eligible for cohort II. 30 of these (97 %) consented (demographics in Table 1). None of 8 ALS patients had clinically definite, 2 (25 %) had clinically probable, 2 (25 %) had clinically probable-laboratory supported, and 4 (50 %) had clinically possible ALS. All 5 MMN patients had definite MMN. 17 (53 %) patients in cohort II were neither diagnosed as ALS nor as MMN, but judged as follows: suspected ALS without fulfillment of revised El-Escorial criteria (3 patients); spinal muscular atrophy (3); cervical myelopathy (2), flail arm syndrome (2); neuralgic shoulder amyotrophy (1); suspected MMN without fulfillment of EFNS/PNS guideline (1), diabetic neuropathy (1); traumatic radiculopathy (1); sensorimotor focal neuropathy (2), and somatoform disorder (1).

More US than NCS measurements showed group differences in both cohort I (Table 2, Online Resource Table 1) and cohort II (Online Resource Tables 2 and 3). Whereas in cohort I only 5/32 (16 %) of NCS measurements showed group differences, this was the case in 15/26

Table 1 Demographics, paraclinical findings and symptoms scores

	Cohort I			Cohort II				
	ALS ($n = 16$)	MMN ($n = 8$)	Test	ALS ($n = 8$)	MMN ($n = 5$)	Other ($n = 17$)	Test	Group differences
Demographics								
Age, mean (SD), years	58.7 (8.2)	69.6 (7.5)	$t(22) = -2.95$, $p = 0.008$[a]	65.1 (8.8)	53.6 (11.7)	60.3 (14.7)	$F(2, 27) = 1.21$, $p = 0.341$[b]	
Female sex, no. (%)	4 (25 %)	2 (25 %)	$p = 1.000$[e]	6 (75 %)	2 (40 %)	17 (29 %)	$p = 0.032$[f]	
Symptom onset, mean (SD) for cohort I, median (IQR) for cohort II, months	78 (125)	111 (71)	$t(20) = -0.72$, $p = 0.480$[a]	14 (22)	36 (82)	39 (56)	$H(2) = 3.24$, $p = 0.198$[d]	
Diagnosis time point, mean (SD), months	22 (14)	56 (27)	$t(19) = -2.62$, $p = 0.039$[a]	n.a.	n.a.	n.a.		
Paraclinical findings								
GM1-IgM antibodies for those tested, *n* (%)	4 (33 %)	4 (50 %)	$p = 0.651$[e]	0 (0 %)	4 (100 %)	9 (82 %)	$p = 0.005$[f]	ALS < MMN; ALS > other
Symptoms								
Handgrip force, mean (SD), NM	7.55 (9.31)	15.14 (9.63)	$t(21) = -1.76$, $p = 0.093$[a]	8.78 (9.77)	25.00 (18.87)	22.94 (12.58)	$F(2, 24) = 2.95$, $p = 0.072$[b]	
MRC-SS, median (IQR)	41.00 (28.00)	55.50 (5.00)	$U = 7.00$, $p = 0.001$[c]	44.75 (6.75)	58.00 (5.75)	56.00 (10.75)	$H(2) = 9.52$, $p = 0.009$[d]	ALS < other

Data are displayed as mean ± SD (normally distributed data) or median (IQR; all other data)

Normality was assessed by Kolmogorov–Smirnov test. For continuous data with normal distribution, two-sided *t* test[a], or ANOVA[b] with post hoc two-sided *t* test was performed. For continuous data without normal distribution and for ordinal data, Mann–Whitney *U* test[c], or Kruskal–Wallis-test[d] with post hoc Mann–Whitney *U* test was performed. For categorical data, Fisher's exact test[e] was performed, in case of three groups with a Freeman–Halton extension[f]. Significant group differences were assumed if $p < 0.05$ for all tests (with Bonferroni adjustment for α inflation)

n number, *SD* standard deviation, *f* female, *m* male, *IQR* interquartile range, *NM* newton meter, *MRC-SS* medical research council symptoms score, *ALS* amyotrophic lateral sclerosis, *MMN* multifocal motor neuropathy, *n.a.* not applicable

(56 %) of US measurements. Figure 1a shows the ROC curves of the three best US and NCS measurement values. The mean AUC of the three best US values was much higher than of the three best NCS values (0.99 ± 0.01 vs. 0.79 ± 0.03, $p < 0.001$). Of note, the six best US measurements were all CSA values; best diameter was from sciatic nerve with an AUC of 0.93 (CI 95 % 0.82–1.00, not shown). There were no adverse events from performing US or NCS measurement.

To study the diagnostic value of measurements from different nerve types (pure sensory, pure autonomous, and sensorimotor nerves), as well as from different body regions (cervical roots vs. peripheral sensorimotor nerves), data were z-transformed and mean *z* scores were calculated from all measurement values of the respective nerve type or body region (Fig. 1b). No group difference was observed for pure sensory or autonomous nerve-derived *z* scores. In contrast, all *z* scores with values from roots or nerves containing motor fibers showed group differences. As the box plots illustrate, mean *z* scores from peripheral sensorimotor nerves separate the groups of MMN and ALS patients better than mean *z* scores from cervical roots. Mean *z* scores from NCS values offered poorer group separation, see also Online Resource Figure 3.

Several strong correlations ($r \geq 0.6$) were found between mean *z* scores of various US and NCS measurement values and clinical scales (MRC-SS, mRS, dynamometer handgrip force, ODSS, r-ALSFR; see Online Resource Table 4 for the strongest correlations found). Figure 2 shows the strongest of the many correlations found between different mean *z* scores of US and NCS measurements and clinical parameters. Whereas correlations between US and clinical parameters could be found in both ALS and MMN groups, correlations between NCS and clinical parameters could be found in the ALS group only.

Various approaches were employed to develop diagnostic models to be tested in cohort II, based on cutoff

Table 2 Group comparisons of nerve ultrasound measurements for cohort I

Nerve	ALS	MMN	Test
Median nerve			
Forearm			
CSA (mm^2)	5.97 (1.06)	11.45 (5.43)	$t(22) = -2.83$, $p = 0.024$[a]
Diameter (mm)	2.33 (0.29)	2.87 (1.00)	$t(22) = -1.51$, $p = 0.171$[a]
Upper arm			
CSA (mm^2)	7.83 (1.72)	19.44 (9.30)	$t(22) = -3.50$, $p = 0.009$[a]
Diameter (mm)	2.56 (0.48)	3.20 (0.79)	$t(22) = -2.10$, $p = 0.063$[a]
Ulnar nerve			
Forearm			
CSA (mm^2)	4.80 (1.29)	8.88 (1.52)	$t(22) = -6.69$, $p < 0.001$[a]
Diameter (mm)	1.93 (0.33)	2.22 (0.44)	$t(22) = -1.77$, $p = 0.090$[a]
Upper arm			
CSA (mm^2)	5.18 (1.07)	7.73 (2.25)	$t(22) = -3.04$, $p = 0.015$[a]
Diameter (mm)	1.88 (0.44)	2.59 (0.51)	$t(22) = -3.51$, $p = 0.002$[a]
Radial nerve			
Upper arm			
CSA (mm^2)	4.00 (1.33)	8.45 (7.40)	$U = 5.00$, $p < 0.001$[b]
Diameter (mm)	1.76 (0.41)	2.33 (2.02)	$U = 13.00$, $p < 0.004$[b]
C5 root			
Inter-scalene gap			
CSA (mm^2)	5.23 (1.84)	11.45 (5.43)	$t(22) = -2.12$, $p = 0.067$[a]
Diameter (mm)	2.24 (0.35)	2.92 (0.75)	$t(22) = -2.44$, $p = 0.039$[a]
C6 root			
Inter-scalene gap			
CSA (mm^2)	7.44 (2.37)	9.23 (3.20)	$t(22) = -1.55$, $p = 0.136$[a]
Diameter (mm)	3.80 (0.69)	4.25 (1.02)	$t(22) = -1.27$, $p = 0.216$[a]
C7 root			
Inter-scalene gap			
CSA (mm^2)	9.88 (2.65)	15.11 (2.73)	$t(15) = -3.95$, $p = 0.001$[a]
Diameter (mm)	3.32 (0.67)	4.03 (1.03)	$t(15) = -1.72$, $p = 0.106$[a]
Superior trunk			
Supraclavicular fossa			
CSA (mm^2)	17.12 (4.22)	28.87 (9.11)	$t(19) = -4.04$, $p = 0.001$[a]
Diameter (mm)	4.84 (0.91)	6.41 (2.15)	$t(18) = -2.26$, $p = 0.036$[a]
Vagal nerve			
Neck			
CSA (mm^2)	1.91 (0.63)	2.07 (0.65)	$t(22) = -0.57$, $p = 0.577$[a]
Diameter (mm)	1.18 (0.30)	1.23 (0.32)	$t(22) = -0.40$, $p = 0.692$[a]
Sciatic nerve			
Upper leg			
CSA (mm^2)	41.88 (8.17)	68.13 (17.08)	$t(18) = -4.05$, $p = 0.003$[a]
Diameter (mm)	5.86 (1.27)	8.01 (1.11)	$t(18) = -3.92$, $p = 0.001$[a]
Tibial nerve			
Lower leg			
CSA (mm^2)	8.26 (1.85)	13.65 (2.52)	$t(20) = -5.76$, $p < 0.001$[a]
Diameter (mm)	2.67 (0.63)	3.62 (0.98)	$t(21) = -2.84$, $p = 0.010$[a]

Table 2 continued

Nerve	ALS	MMN	Test
Sural nerve			
Lower leg			
CSA (mm^2)	1.81 (0.67)	2.01 (0.75)	$t(21) = -0.67$, $p = 0.510$[a]
Diameter (mm)	0.98 (0.26)	1.12 (0.24)	$t(21) = -1.31$, $p = 0.203$[a]

Significant group differences were assumed if $p < 0.05$ for all tests

[a] Normality was assessed by Kolmogorov–Smirnov test. For data with normal distribution, table shows mean and standard deviation (*SD*), as well as a two-sided *t* test

[b] For data without normal distribution, table shows median and interquartile range (*IQR*), as well as a Mann–Whitney *U* test

ALS amyotrophic lateral sclerosis, *MMN* multifocal motor neuropathy, *n* number, *CSA* cross-sectional area

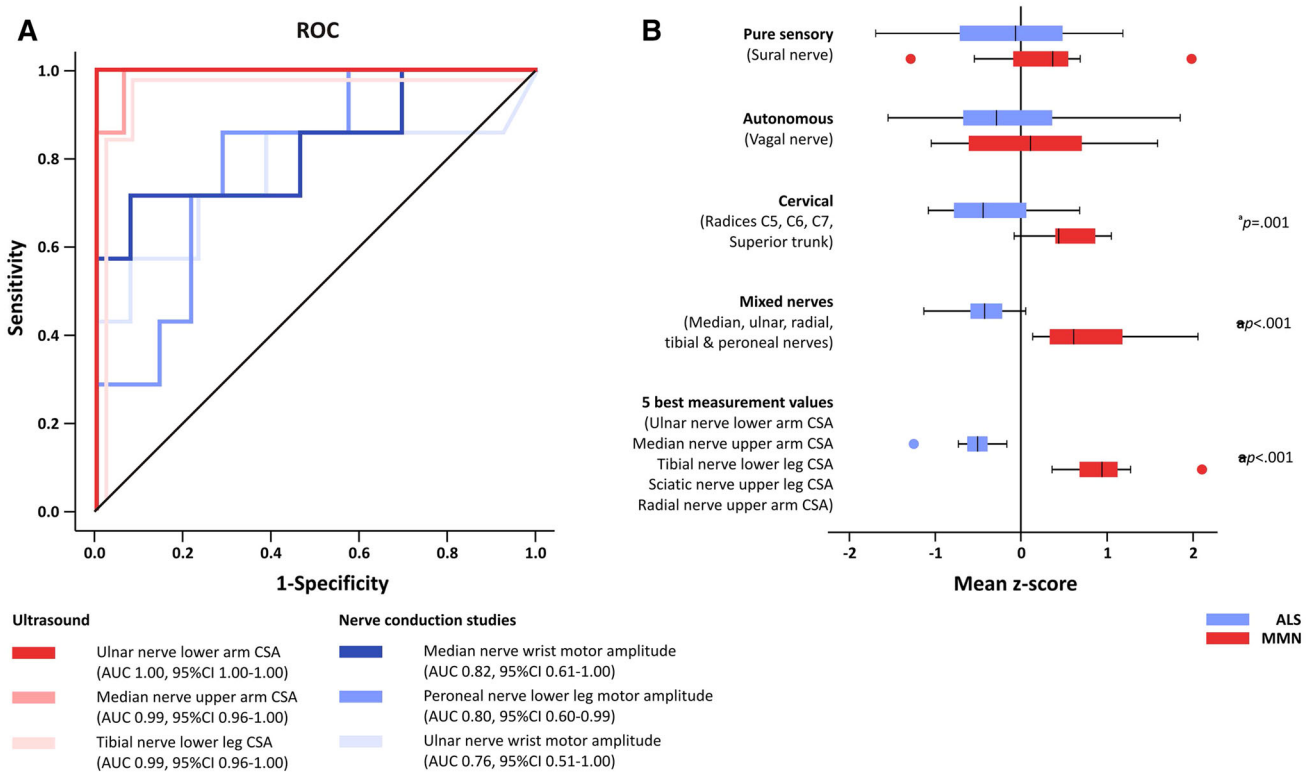

Fig. 1 Comparison of US and NSC for differential diagnosis of ALS/LMND and MMN. **a** ROC curves of three best measurement values from ultrasound (*red curves*) and nerve conduction studies (*blue curves*) in cohort I. **b** Mean *z* scores of ultrasound measurement values from various nerve types (pure sensory and autonomous nerves, cervical roots, peripheral sensorimotor nerves), and from the five measurement values with the highest AUC in ROC analysis in cohort I are shown (ALS/LMND in *blue*, MMN in *red*). Significant group differences (two-sided *t* test) were only found for *z* scores incorporating motor fibers. *Boxes* represent the interquartile range, the *horizontal line* the median, and the antennas the range excluding outside values (defined as values beyond lower/upper quartile [1.5 times interquartile range, shown as *circles*)]. *ROC* receiver operating curve, *AUC* area under the curve, *CSA* cross-sectional area, *95 % CI* 95 % confidence interval, *ALS* amyotrophic lateral sclerosis, *MMN* multifocal motor neuropathy

values defined in the ROC analyses in cohort I. Table 3 shows the diagnostic performance of the US models in terms of sensitivity and specificity for MMN if patients with other diagnoses (other) were excluded (column ALS vs. MMN) or included (column ALS + other vs. MMN, see Online Resource Figures 1 and 2). Even if there was no preselection and the group other was included, the best US model still showed a specificity of 92 % for MMN (Table 3), whereas the best NCS model reached a specificity of only 52 % (Online Resource Table 5). Model US1 (Table 3) provided perfect group separation if only ALS and MMN subjects were considered. The same model

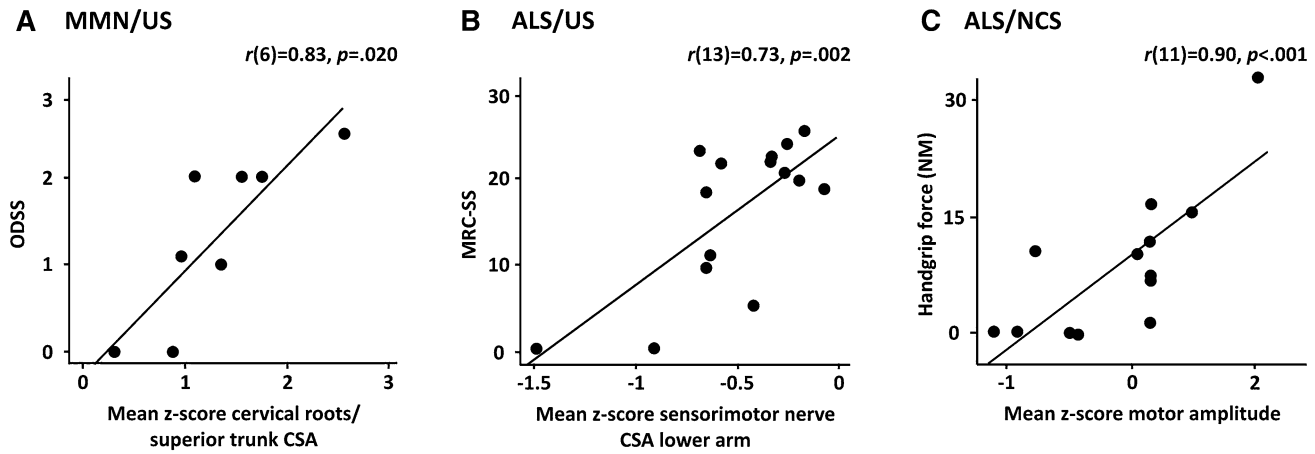

Fig. 2 Correlations between clinical scales and mean *z* scores of ultrasound and nerve conduction studies measurement values in cohort I. Strongest correlations between mean *z* scores of US (**a**, **b**) and NCS (**c**) measurement values and clinical scales ODSS (**a**), MRC sum score (**b**), and handgrip force (**c**) in cohort I. Pearson's correlation coefficient is displayed in the *upper right part* of each graph. Displayed are the strongest correlations between the respective patient group and diagnostic method only, in the case that more than one strong correlation was found. No correlation was found for all tested NCS mean *z* scores in MMN patients. Of the 112 combinations tested for US values, 26 (23 %) showed significant ($p < 0.05$) correlations. Of the 136 NCS combinations tested, only 13 (10 %) showed significant correlations. Of the 26 significant US correlations, 18 (70 %) were strong ($r \geq 0.60$) and 8 (30 %) moderate ($r = 0.30$–0.59). Of the 13 significant NCS correlations, 11 (85 %) were strong and 2 (15 %) moderate (see Online Resource Table 4 for complete list of correlation analyses and their statistical results). *MMN* multifocal motor neuropathy, *US* ultrasound, *CSA* cross-sectional area, *ALS* amyotrophic lateral sclerosis, *NCS* nerve conduction studies, *NM* Newton meter

produced only two false-positives for MMN (9 %) if patients with other diagnoses than ALS or MMN were not excluded from cohort II. Table 4 provides the cutoff values needed for the application of this most efficient US1 model. In contrast, best NCS model (NCS3, Online Resource Table 4) showed a much poorer performance with one false-positive for MMN (13 %) if only ALS and MMN subjects were included, but 14 (64 %) false-positives for MMN in the whole cohort II. Since study groups were unbalanced for some baseline characteristics (age, gender, and disease duration), their relevance as covariates for the observed group differences was tested in generalized linear models (GLM). For the sciatic nerve only (7.69 % of all US measurements) there was a significant effect ($p < 0.05$, for CSA: age and disease duration, for diameter: disease duration). However, the sciatic nerve was not included in any model suggested for diagnostic usage (Table 4).

Discussion

Particularly at early disease stages the differential diagnosis of ALS/LMND and MMN remains a challenge [2, 4, 5]. At the same time, correct differential diagnosis is of high therapeutic and prognostic importance. Our study provides evidence that peripheral nerve US reliably distinguishes the two diseases. This is illustrated by very high AUC values of single US measurement values and very high correct classification rates in a prospectively and consecutively recruited confirmation cohort of hospital-admitted patients (cohort II), reaching 100 % sensitivity and 92–100 % specificity for detecting MMN, depending on the exclusion of other diagnoses.

The study further indicates that US might be superior to NCS in the differential diagnosis of ALS/LMND and MMN as a single tool: First, more individual US than NCS values showed significant group differences, as well as higher AUC values. Second, the resulting diagnostic NCS models had poorer reclassification rates for MMN in cohort II, particularly if not only ALS/LMND and MMN patients, but all consecutive patients including those with other diagnoses were analyzed. It is even likely that our study overestimated the diagnostic quality of NCS. Current diagnostic criteria for MMN include NCS motor conduction block as a major criterion. Since our reference standard (EFNS/PNS guideline) included this criterion and conduction blocks were at the same time included in the diagnostic NCS models, our study was possibly biased in favor of NCS because of its double role both as a diagnostic method to be tested and in the definition of the reference standard. Of note, one of the patients misclassified by US in cohort II failed the diagnostic criteria for possible MMN because of the lack of a motor conduction block in NCS. If there were diagnostic criteria for both ALS and MMN not relying on NCS, then our comparison

Table 3 Five best ultrasound models and diagnostic performance in cohort II

Model no.	Measurement values selection	ROC cutoff selection	No. of values	Measurement values	No. of path. values	Cohort II ALS vs. MMN		Cohort II ALS + other vs. MMN	
						Specificity (%)	Sensitivity (%)	Specificity (%)	Sensitivity (%)
US1	CSA only, ROC AUC ≥0.9	90 % specificity	8	CSA: Ulnar n. (LA), Medial n. (LA&UA), Radial n. (UA), Superior Trunk, C7, Sciatic n. (UL), Tibial n. (LL)	3	100	100	92	100
US2	CSA and diameter, ROC AUC ≥0.9	90 % specificity	9	CSA: Ulnar n. (LA), Medial n. (LA&UA), Radial n. (UA), Superior Trunk, C7, Sciatic n. (UL), Tibial n. (LL), Diameter: Sciatic n. (UL)	3	100	100	92	100
US3	CSA and diameter, ROC AUC 95 % CI >0.5	80 % specificity	16	CSA: Ulnar n. (LA&UA), Medial n. (LA&UA), Radial n. (UA), Superior Trunk, C5, C6, C7, Sciatic n. (UL), Tibial n. (LL), Diameter: Ulnar n. (UA), Radial n. (UA), C7, Sciatic n. (UL), tibial n. (LL)	6	100	100	84	100
US4	CSA only ROC AUC 95 % CI >0.5	80 % specificity	11	CSA: Ulnar n. (LA&UA), Medial n. (LA&UA), Radial n. (UA), Superior Trunk, C5, C6, C7, Sciatic n. (UL), Tibial n. (LL)	5	100	100	84	100
US5	CSA only ROC AUC 95 % CI >0.5	90 % specificity	11		4	100	100	80	100

The five best diagnostic ultrasound models found by the approach as described in the text are shown. Column "measurement values selection" indicates the criteria for the inclusion of values into the model: for e.g., for model "US1" only CSA values with a diagnostic accuracy of ≥0.9 in ROC AUC were included. Column "cut-off selection" indicates the criteria for the pathological cutoff value. Column "no. of values" gives the number of single measurement values included. Column "measurement values" names the included measurement values. Column "no. of pathological values" indicates how many of the measured values have to be above the pathological cutoff to reach the diagnostic accuracy as indicated. "ALS vs. MMN" indicates the diagnostic accuracy in cohort II if only ALS and MMN patients are regarded. "ALS + other vs. MMN" shows the diagnostic accuracy for the contrast between MMN patients on the side and all other patients with either ALS or other causes in cohort II

No. number, *ROC* receiver operating curve, *path.* pathological, *ALS* amyotrophic lateral sclerosis, *MMN* multifocal motor neuropathy, *CSA* cross-sectional area, *AUC* area under the curve, *n.* nerve, *LA* lower arm, *UA* upper arm, *UL* upper limb, *LL* lower limb, *95 % CI* 95 % confidence interval

of the two methods would have been more balanced and the results might have been even more in favor of US.

The requirement of only 8 measurements with an approximate total examination time of 20 min for the optimized US model (US1) documents the feasibility of US in routine clinical diagnostic workup.

The results of this study are in line with previous investigations on US in MMN and ALS. Beekman and colleagues showed that in MMN multiple nerve enlargements could be found along the entire course of motor nerves [10]. Despite MMN being considered a focal neuropathy leading to the electrophysiological hallmark of motor conduction blocks, US nerve enlargements could be found independent from pathological NCS results and even in nerves with no clinical affection. This disperse character of US changes in MMN provides a rationale for using fixed and reproducible US examination points in all patients in our study instead of searching for focal enlargements along the entire course of nerves. The finding that such a simple and reproducible examination protocol results into very good group separation is an important presupposition for routine clinical application. The US changes of ALS (decrease in nerve caliber) oppose those of MMN, possibly due to axonal degeneration as a consequence of LMN disease [14, 21].

Table 4 CSA measurements for model US1 and respective pathological cutoff values

Measurement (CSA)	ROC AUC (95 % CI)	Cutoff value 90 % specificity (mm^2)
Ulnar nerve lower arm	1.00 (1.00–1.00)	≥6.05
Median nerve upper arm	0.99 (0.96–1.00)	≥9.83
Tibial nerve lower leg	0.99 (0.96–1.00)	≥10.33
Sciatic nerve upper leg	0.98 (0.92–1.00)	≥55.77
Radial nerve upper arm	0.95 (0.87–1.00)	≥5.63
Median nerve lower arm	0.94 (0.83–1.00)	≥7.53
C7	0.92 (0.77–1.00)	≥12.47
Superior trunk	0.90 (0.77–1.00)	≥23.33

US measurement values included in the US model US1 (see Table 3) with the best proportion between measurement efficacy and diagnostic accuracy. The respective ROC AUC with 95 % confidence interval is shown, as well as the respective cutoff value with 90 % specificity. All values included in the model are CSA values

CSA cross-sectional area, *ROC* receiver operating curve, *path.* pathological, *AUC* area under the curve, *95 % CI* 95 % confidence interval

Another study on the role of US in the differential diagnosis of ALS and MMN was published recently, but chose a different approach to derive with a diagnostic model and did not provide a direct comparison with NCS performance [15]. Instead of defining specific cutoffs for single US measurements in a defined patient group of ALS and MMN patients (cohort I in our study), this study relied on previously published cutoff values defined in other disease contrasts than the ALS vs. MMN contrast. In addition, the measurement protocol did not use osseous reference points as in our study (having the advantage of being easily reproducible) [20]. Four of the eight measurement values in our best US model (US1, Table 3) were not assessed in the afore-mentioned study (radial nerve, superior trunk, C7, sciatic nerve). All these aspects might explain the lower diagnostic accuracy of 87.5 % sensitivity and 94.1 % specificity in comparison to the models presented in our study.

Significant group differences were only found for mean *z* scores from nerves containing motor fibers. Additionally, mean *z* scores from distal nerve segments provided better group separation than cervical nerve roots. The best single AUC of 1.00 (95 % CI 1.00–1.00) was found for ulnar nerve CSA at the forearm, but other measurement sites had very similar AUC values (Fig. 1a).

Our study has several limitations. First, it was a monocenter study with a restricted number of participants. Even though patients were transferred from all district hospitals with a Department of Neurology in the City of Dresden (>530,000 inhabitants) and from nearby regions of east Saxony (>1,000,000 inhabitants), only 5 MMN patients could be recruited for cohort II. Due to this low subject number, further subgroup analyses, for e.g., of patients at early disease stages in cohort II, were not carried out. A multicenter validation study is thus warranted to confirm the discriminatory power of US for ALS vs. MMN in a routine clinical setting. Second, as already mentioned, NCS was both an index test, of which the diagnostic quality was assessed, and a component of the reference standard. Thus, our results are likely to be biased in favor of NCS. Third, all US examinations were performed by two experienced examiners. However, the applied examination protocol with a limited number of well-defined examination spots should facilitate its application on a broader scale.

Clinical implications

Nerve US is of high diagnostic quality in the differential diagnosis of ALS/LMND and MMN. It may be superior to NCS in the diagnosis of MMN in hospital-admitted patients with a suspected diagnosis of ALS/LMND or MMN.

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Compliance with ethical standards

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