European Journal of Neurology

european journal of neurology
the official journal of the european academy of neurology

ORIGINAL ARTICLE OPEN ACCESS

Split Hand Syndrome in Charcot–Marie–Tooth Disease Type X1 (CMTX1): A Clinical, Neurophysiological, and Radiological Study

Alessandro Bertini[1] | Marco Moscatelli[2,3] | Claudia Ciano[4] | Mattia Verri[2] | Eleonora Cavalca[1] | Luca Maria Sconfienza[3,5] | Marina Grisoli[2] | Paola Lanteri[4] | Davide Pareyson[1] | Chiara Pisciotta[1] | the Italian CMT Network

[1]Unità Malattie Neurologiche Rare, Dipartimento di Neuroscienze Cliniche, Fondazione IRCCS Istituto Neurologico Carlo Besta, Milan, Italy | [2]Unità di Neuroradiologia, Dipartimento di Diagnostica e Tecnologia, Fondazione IRCCS Istituto Neurologico Carlo Besta, Milan, Italy | [3]Dipartimento di Scienze Biomediche per la Salute, Università di Milano, Milan, Italy | [4]Unità di Neurofisiopatologia, Dipartimento di Diagnostica e Tecnologia, Fondazione IRCCS Istituto Neurologico Carlo Besta, Milan, Italy | [5]IRCCS Istituto Ortopedico Galeazzi, Milan, Italy

Correspondence: Chiara Pisciotta (chiara.pisciotta@istituto-besta.it)

Received: 27 January 2025 | **Revised:** 9 April 2025 | **Accepted:** 27 April 2025

Funding: This work was partially funded by Telethon-Italy Foundation grant GUP13006 and partially supported by the Italian Ministry of Health (RRC).

Keywords: Charcot–Marie–tooth disease | CMTX1 | split hand

ABSTRACT

Background: Split hand syndrome (SHS) is a hand atrophy pattern characterized by predominant wasting in the thenar muscles (abductor pollicis brevis-APB, first dorsal interosseous-FDI), with relative sparing of the hypothenar (abductor digiti minimi-ADM). SHS was also reported in CMTX1, but eventually attributed to median–ulnar dissociated involvement. We investigated the presence and specificity of SHS in CMTX1.

Methods: We gathered clinical/neurophysiological/radiological information in CMTX1 and non-CMTX1 patients, including disease severity (CMT Examination Score-CMTES) and compound muscle amplitude potential (CMAP) of APB/FDI/ADM. We obtained ADM/APB ratio (ADM/APBr) = $CMAP^{ADM}/CMAP^{APB}$ and split-hand index (SHI) = $(CMAP^{APB} \times CMAP^{FDI})/CMAP^{ADM}$. Eight patients underwent 3T MRI of the hand muscles. We defined SHS based on three criteria: clinical, neurophysiological, and radiological, with at least one criterion required for SHS diagnosis. Through ADM/APBr > 1.7, we assessed the specificity of SHS for CMTX1 among the Italian CMT Registry cohort, encompassing 750 clinically well-characterized patients.

Results: We evaluated 22 CMTX1 (age 41.3 ± 12.2) and 40 non-CMTX1 (49.2 ± 14.9). 50% (vs. 3% of non-CMTX1, $p < 0.001$) and 64% (vs. 0%, $p < 0.001$) of CMTX1 had clinical and neurophysiological SHS, respectively. In CMTX1, SHS was independent from gender and hand dominance. Both ADM/APBr and SHI correlated with disease duration (rs = 0.77, $p < 0.001$; rs = −0.65, $p = 0.022$, respectively), and CMTES (rs = 0.56, $p = 0.006$; rs = −0.62, $p < 0.001$, respectively). 3/8 CMTX1 had radiological SHS. Within the Italian CMT Registry cohort, ADM/APBr > 1.7 yielded a specificity of 74% for CMTX1 females among I-CMT/CMT2, and 85% for CMTX1 males among CMT1/I-CMT.

Discussion: Up to 59% of CMTX1 patients develop thenar-hypothenar (rather than median-ulnar) SHS. MRI is a novel approach to detect this dissociated denervation pattern. ADM/APBr > 1.7 may be useful to address *GJB1* testing in males.

See Data S1 for the members of the Italian CMT Network.

1 | Introduction

The split hand syndrome (SHS) is a dissociated atrophy pattern in hands, characterized by predominant wasting in the thumb side (thenar, to simplify) muscles, namely abductor pollicis brevis (APB) and first dorsal interosseous (FDI), with relative sparing of the abductor digiti minimi (ADM) in the hypothenar eminence [1, 2]. This clinical pattern does not respect root or nerve trunk territories since all involved muscles are innervated by C8–T1 [3], with FDI and ADM being both innervated by the ulnar nerve and APB by the median nerve.

SHS was first described in amyotrophic lateral sclerosis (ALS) [4, 5]. Different neurophysiological criteria for SHS have been proposed, with the aim of prompting early diagnosis (rule in rather than rule out) in patients with ALS. The ADM/APB ratio (ADM/APBr) and split-hand index (SHI) are among the most used ones. The ADM/APBr is calculated by dividing the compound muscle amplitude potential (CMAP) amplitude of the corresponding muscles. Conversely, the SHI is derived as follows: $SHI = (CMAP^{APB} \times CMAP^{FDI})/CMAP^{ADM}$. According to previous studies, ADM/APBr values > 1.7 and SHI values < 5.2 are considered abnormal, yielding 95–99% [1, 6, 7] and 80% [1, 8, 9] of specificity for ALS, respectively, when compared with mimic disorders or healthy controls.

Over the years, SHS has gradually lost its specificity for ALS, being described in other motor neuronopathies such as Spinal and Bulbar Muscular Atrophy (SBMA) [9] and Spinal Muscular Atrophy [10].

Interestingly, in 2017, Tomaselli and colleagues [11] found that 48% (12/25) of Charcot–Marie–Tooth disease type X1 (CMTX1) patients in their series showed clinical SHS, which was eventually attributed to median-ulnar (with median territory more involved) dissociated atrophy, rather than thenar-hypothenar. This is noteworthy as CMTX1 is a demyelinating/intermediate, length-dependent neuropathy caused by variants in the *GJB1* gene, clinically and pathogenetically extremely different from motor neuronopathies.

Objective of the study was to explore the presence of SHS in patients with CMTX1compared to other CMT subtypes through clinical, neurophysiological, and radiological evaluation.

Subsequently, we assessed the specificity of SHS for CMTX1 among all CMTs in a broader cohort, taking advantage of the Italian CMT Registry [12].

Importantly, throughout all the study, we focused on the originally reported thenar-hypothenar dissociation, to eventually assess whether CMTX1 behaves similarly to motor neuronopathies as far as hand atrophy pattern is concerned. This can potentially shed light on the role of the peripheral nervous system (PNS) in the genesis of SHS, and have important implications for CMT diagnosis (if SHS proves typical of CMTX1 among CMTs) and management.

2 | Methods

2.1 | Aim 1. Frequency of SHS in CMTX1

2.1.1 | Cohort

We gathered a minimal dataset of clinical and neurophysiological information in clinically symptomatic CMTX1 patients and genetically confirmed non-CMTX1 patients recruited among other most frequent subtypes (CMT1A, CMT1B, CMT2A, CMT2F, CMT2I/J, CMT4C), including current age, hand dominance, disease severity (CMT Examination Score-CMTES) [13], disease onset (age at first motor or sensory symptom), disease duration, and strength (Medical Research Council-MRC) in APB, FDI, and ADM muscle pairs.

To minimize false positives, patients with all three (APB, FDI, ADM muscles) CMAPs < 1 mV, symptoms consistent with carpal tunnel syndrome (CTS), and/or history of previous CTS surgery, were not included.

2.1.2 | Clinical SHS

Clinical SHS was defined by the presence of both [ΔMRC (ADM vs. FDI) ≥ 1 AND ΔMRC (ADM vs. APB) ≥ 1]. We considered $4- = 3.75$ and $4+ = 4.25$. (Table 1).

2.1.3 | Neurophysiological SHS

The CMAPs from the thenar (APB and FDI) and hypothenar (ADM) muscles were evoked by supramaximal stimulation of the median and ulnar nerves at the wrist. Skin temperature was maintained at 34°C, and limbs were warmed if necessary. CMAP amplitudes were measured peak-to-peak.

TABLE 1 | Split hand syndrome criteria.

SHS type	Criteria	Patients (*N*)
Clinical SHS	[ΔMRC (ADM vs. FDI) ≥ 1 AND ΔMRC (ADM vs. APB) ≥ 1]	22
Neurophysiological SHS	[(ADM/APBr > 1.7) AND (SHI < 5.2)]	14
Radiological SHS	• [FF (APB AND FDI at least doubled as compared to ADM)] OR • [ΔAtrophy Score (APB vs. ADM) AND (FDI vs. ADM) ≥ 1]	8

Abbreviations: ADM, abductor digiti minimi; ADM/APBr, ADM/APB ratio; APB, abductor pollicis brevis; FDI, first dorsal interosseous; FF, fat fraction; *N*, number; SHI, split hand index; SHS, split hand syndrome.

Neurophysiological SHS was defined by the presence of both [(ADM/APBr > 1.7) AND (SHI < 5.2)]. (Table 1).

2.1.4 | Radiological SHS

In a few CMTX1 patients, hand muscles were studied bilaterally on a 3T MRI scanner (Phillips Achieva) with STIR and T1 3D Dixon sequences. APB, FDI, and ADM muscle involvement was graded by a blinded radiologist with long experience in neuromuscular disorders based on three parameters: (a) degree of fat infiltration assessed with Fat Fraction (FF, by drawing ROIs on water images); (b) degree of muscle atrophy assessed with a semi-quantitative Atrophy Score from 0 to 4 (0 = normal; 4 = severe atrophy); (c) the presence of intra-muscular T2 hyperintensity in STIR.

Radiological SHS was defined by [FF (APB AND FDI at least doubled as compared to ADM)] OR [ΔAtrophy Score (APB vs. ADM) AND (FDI vs. ADM) ≥ 1]. (Table 1).

2.1.5 | Definition of SHS

We defined SHS when at least one of the three criteria was met: clinical, neurophysiological, or radiological.

2.2 | Aim 2. Specificity of SHS for CMTX1 Among All CMTs

Afterwards, we took advantage of ADM/APBr > 1.7 neurophysiological criterion to retrospectively assess the specificity of SHS for CMTX1 among all CMTs in the large Italian CMT Registry cohort, which encompasses 1012 CMT patients from 9 centres, as previously described by Pisciotta and colleagues [12]. The advantage of ADM/APBr compared to SHI is that the ulnar nerve is derived from ADM in most patients.

For Aim 2, we excluded patients with both APB and ADM muscles CMAPs < 1 mV, those with a history of previous CTS surgery, and subjects diagnosed with Hereditary Neuropathy with Liability to Pressure Palsies (HNPP), caused by *PMP22* deletion.

Data were pseudo-anonymized. The Institutional Ethics Committee of INB (10 November 2021, no. 89/2021, and 07 February 2018, no. BST 12/2018) approved the study. All participants gave informed consent.

2.3 | Statistical Analysis

A description of participant characteristics at baseline was provided in terms of absolute numbers and percentages for categorical data, and means ± standard deviations (range), or medians with Interquartile Range (IQR) for continuous data, as appropriate. We used the Mann–Whitney U Test and Fisher's exact test, as appropriate, to compare CMTX1 with non-CMTX1 patients (Aim 1). We used Spearman's 'rho' correlation to detect the association of SHI and ADM/APBr with CMTES and disease duration. Throughout the statistical analysis, the significance level was set at 0.05 (significant if $p < 0.05$). The p value was rounded to three decimal places; p values lower than 0.001 were expressed as $p < 0.001$.

3 | Results

3.1 | Aim 1. Frequency of SHS in CMTX1

Overall, we recruited 22 right-handed CMTX1 (13 males; mean age 41.3 ± 12.2, range 19–67) and 40 right-handed non-CMTX1 patients (13 males; mean age 49.2 ± 14.9, range 20–80), evaluating 44 and 58 hands, respectively. Among 40 patients with non-CMTX1, 22 had CMT1A (*PMP22* duplication), 3 had CMT1B (*MPZ*), 5 had CMT2A (*MFN2*), 5 had CMT2F (*HSPB1*), 4 had CMT2I/J (*MPZ*), and 1 had CMT4C (*SH3TC2*). Disease severity (CMTES) (9.8 ± 3.6 vs. 9.2 ± 5.0, $p = 0.703$) and duration (23.7 ± 10.9 vs. 23.8 ± 16.8, $p = 0.706$) were similar between CMTX1 and non-CMTX1 patients.

For details on clinical, neurophysiological, and radiological information on recruited patients, see Table 2 (CMTX1) and Table S1 (non-CMTX1).

We found that 50% (11/22; bilateral in 10/11, and unilateral in 1/11) of CMTX1 patients showed clinical SHS (Figures 1 and 2). Conversely, of non-CMTX1, only one (3%, 1/36) patient (with CMT2I/J) showed a dissociated atrophy pattern in hands ($p < 0.001$).

Remarkably, neurophysiological SHS was exclusive of CMTX1 patients, being detected in 64% (9/14 vs. 0/16; $p < 0.001$) of cases, and in 65% (17/26 vs. 0/32; $p < 0.001$) of hands. Two out of 9 patients with neurophysiological SHS were asymptomatic for clinical SHS. As far as CMTX1 is concerned, both disease duration and severity (CMTES) strongly correlated with neurophysiological SHS (for both SHI: inverse correlation; rs = −0.65, $p = 0.022$; rs = −0.62, $p < 0.001$ respectively; and ADM/APBr: direct correlation; rs = 0.77, $p < 0.001$; rs = 0.56, $p = 0.006$, respectively) (Figure 3). All patients with disease duration > 14 years from first symptom, or CMTES > 10, which reflects a moderate burden of disease, had neurophysiological SHS. ADM/APBr strongly correlated with SHI in our CMTX1 cohort (rs = −0.92, $p < 0.001$).

Interestingly, despite showing lower disease burden as compared to males, due to the casual X chromosome inactivation [14, 15], CMTX1 females were not less prone to develop both clinical (44% vs. 54%, $p = 1$) and neurophysiological (67% vs. 63%, $p = 1$) SHS as compared to males.

Notably, in our right-handed CMTX1 cohort, we could not find an association with hand dominance (mean right SHI = 3.6 ± 5.9 vs. left SHI = 4.4 ± 5.8, $p = 0.701$; median right ADM/APBr = 2.9, IQR 9.18 vs. left ADM/APBr = 2.2, IQR = 18.6 $p = 0.719$).

Eight patients performed bilateral hand muscle MRI (Table 3 and Figure 4). Of them, 38% (3/8) had bilateral radiological SHS. Moreover, 63% (5/8) of patients showed STIR hyperintensity in APB and/or FDI muscles but not in ADM.

TABLE 2 | Clinical and neurophysiological data of patients with CMTX1.

Patient number	Gender	CMTES	Disease duration (years)	MRC APB/ FDI/ADM R	MRC APB/ FDI/ADM L	CMAP (mV) APB/ FDI/ADM R	CMAP (mV) APB/ FDI/ADM L	ADM/ APBr R/L	SHI R/L
1	F	2	2	4+/4/4+	4+/4/4	19.9/12.5/12.4	16.7/11/12.5	0.62/0.75	20.06/14.70
2	F	7	25	4+/4/4+	4+/4/4	4.2/NA/3.8	2.2/NA/2.7	0.90/1.23	NA/NA
3	F	10	37	3/3/3	0/3/3	3.2/NA/12.1	1.5/9/7.6	3.8/**5.07**	NA/**1.78**
4	F	12	34	**0/2/4−**	**0/2/4−**	**0.4/1.1/5.0**	**0.4/0.3/5.6**	**12.50/14.00**	**0.09/0.02**
5	F	11	33	**0/2/3**	**0/2/3**	**0.1/1.0/5.2**	**0.4/0.5/5.3**	**52.00/13.30**	**0.02/0.03**
6	F	17	48	**1/1/3**	3/3/3	**2.0/4.0/8.1**	**1.3/3.5/9.9**	**4.05/7.62**	**0.99/0.46**
7	F	NA	10	4+/4−/4+	4+/4/4+	12.0/5.6/9.8	15.3/6.9/9.1	0.82/0.59	6.86/11.6
8	M	5	14	4+/4+/4+	4/4+/4	9.1/7.9/12.0	16.4/10.9/12.3	1.32/0.75	5.99/14.5
9	M	6	15	4−/4+/4	4+/4+/4	4.0/11.6/6.1	10.4/8.3/8.7	1.53/0.84	7.61/9.92
10	M	8	NA	**0/1/3**	**0/1/2**	NA/NA/NA	NA/NA/NA	NA/NA	NA/NA
11	M	9	14	3/4−/4+	3/4/4+	**3.5/2.6/6.4**	**2.4/5.1/5.3**	**1.83/2.20**	**1.42/2.30**
12	M	9	15	**1/1/3**	**1/1/3**	NA/NA/NA	NA/NA/NA	NA/NA	NA/NA
13	M	9	27	**1/1/4**	**3/2/4**	NA/NA/NA	3.2/NA/7.1	NA/2.21	NA/NA
14	M	11	25	**0/2/3**	**0/2/3**	**1.7/1.6/5.0**	**0.6/1.1/3.7**	**2.94/6.17**	**0.54/0.18**
15	M	12	27	1/3/4−	1/3/4−	NA/NA/NA	NA/NA/NA	NA/NA	NA/NA
16	M	12	31	**0/1/3**	**0/1/3**	**0.1/0.3/2.7**	**0.3/0.7/3.8**	**27.00/12.67**	**0.01/0.06**
17	M	13	28	**1/1/4−**	**1/1/3**	**0.1/1.8/7.1**	**0.5/3.7/7.7**	**71.00/14.40**	**0.03/0.24**
18	M	14	NA	4+/4+/5	4+/4+/5	7.1/NA/5.5	4.5/6.7/4.8	NA/1.07	NA/6.3
19	M	15	24	**0/2/3**	**0/2/3**	**0.1/0.2/1.1**	**0.2/0.3/1.2**	**11.00/6.0**	**0.02/0.05**
20	F	8	10	**0/2/3**	**0/2/3**	NA/NA/NA	NA/NA/NA	NA/NA	NA/NA
21	M	8	26	4−/4−/4−	4−/4−/4−	NA/NA/NA	NA/NA/NA	NA/NA	NA/NA
22	F	7	28	3/4/4	4+/4+/4+	9.6/NA/13.0	NA/NA/NA	1.35/NA	NA/NA/NA

Note: Highlighted in bold clinical and neurophysiological SHS (see Section 2).
Abbreviations: ADM, abductor digiti minimi; APB, abductor pollicis brevis; CMAP, compound muscle action potential; CMTES, CMT Examination score; F, female; FDI, first dorsal interosseus; L, left; M, male; MRC, Medical Research Council (MRC) Scale; NA, not available; R, right.

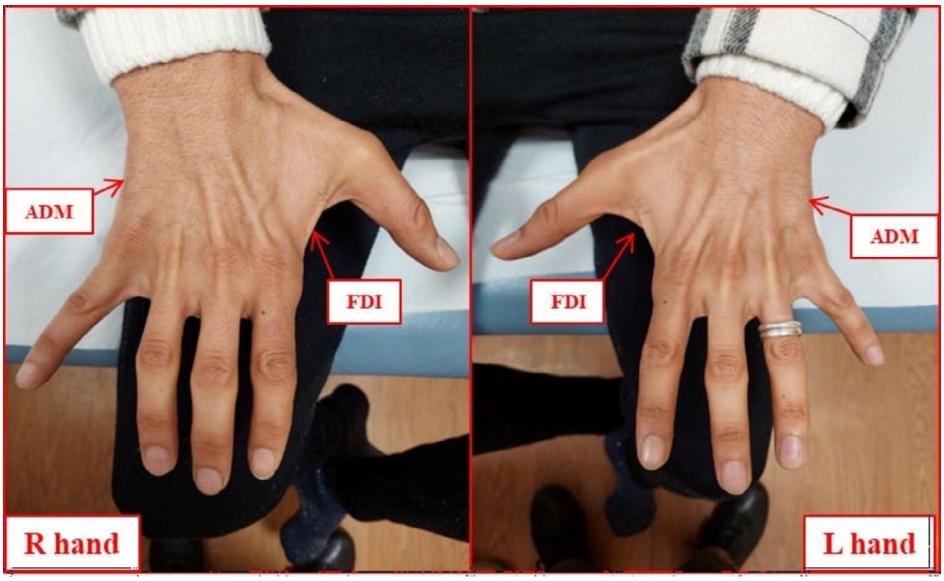

FIGURE 1 | Clinical split hand syndrome in a patient with CMTX1. A male patient (patient 17, see Table 2) with CMTX1 while abducting fingers, showing a thenar-to-hypothenar gradient of atrophy and weakness, with the ADM (MRC R/L = 4−/3) muscle being relatively spared as compared to FDI (MRC R/L = 1/1). ADM, Abductor digiti minimi muscle; FDI, First dorsal interosseous; L, left; R, right.

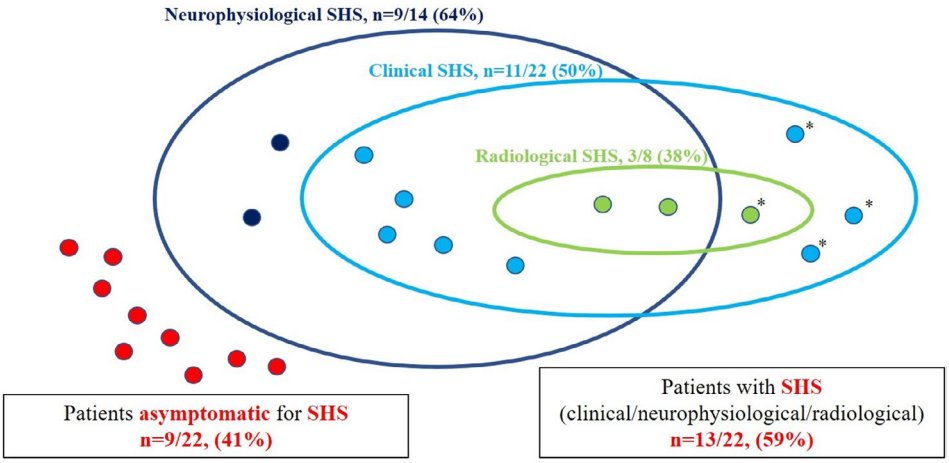

FIGURE 2 | Overview of SHS presence in our CMTX1 cohort. *n*, number; SHS, Split hand syndrome. *Neurophysiological evaluation not available.

Taken together, up to 59% (13/22) of CMTX1 in our whole cohort had SHS, either clinical, neurophysiological, or neuroradiological. (Figure 2).

3.2 | Aim 2. Specificity of SHS for CMTX1 Among All CMTs

Afterwards, we assessed the specificity of SHS for CMTX1 in a larger cohort of CMT patients through ADM/APBr > 1.7 (Figure 5). We took advantage of data collected retrospectively into the Italian CMT Registry: [12] clinical information was available in 750/1012 (74%) of enrolled patients, while both right-hand median and ulnar CMAPs were recorded from APB and ADM, respectively, in 321/750 (43%). We excluded 26 patients with both APB and ADM CMAPs not recordable, 22 with CTS, and 21 with HNPP. Two hundred fifty-two (252; 43 CMTX1) patients were eventually included: 163 had CMT1, 65 had CMT2, and 24 had intermediate-CMT (I-CMT). We assessed that ADM/APBr > 1.7 had a specificity for CMTX1 of 81% (CI 75–86%) among all CMTs.

According to previous large cohort studies on CMTX1, females show nerve conduction velocities in the intermediate-to-axonal range, entering into differential diagnosis with I-CMT/CMT2, while males usually have slowed nerve conduction velocities in the intermediate range [16, 17], falling into differential diagnosis with CMT1/I-CMT. Thus, we evaluated males and femalesseparately: ADM/APBr > 1.7 had a specificity of 74% (CI 56–87%) for CMTX1 females among all I-CMT/CMT2 in our cohort, and of 85% (CI 78–90%) for CMTX1 males among CMT1/I-CMT (Figure 5).

4 | Discussion

In this work, we extensively explored the presence and frequency of SHS in CMTX1. Subsequently, we assessed the specificity of SHS for CMTX1 in a large cohort of CMTs.

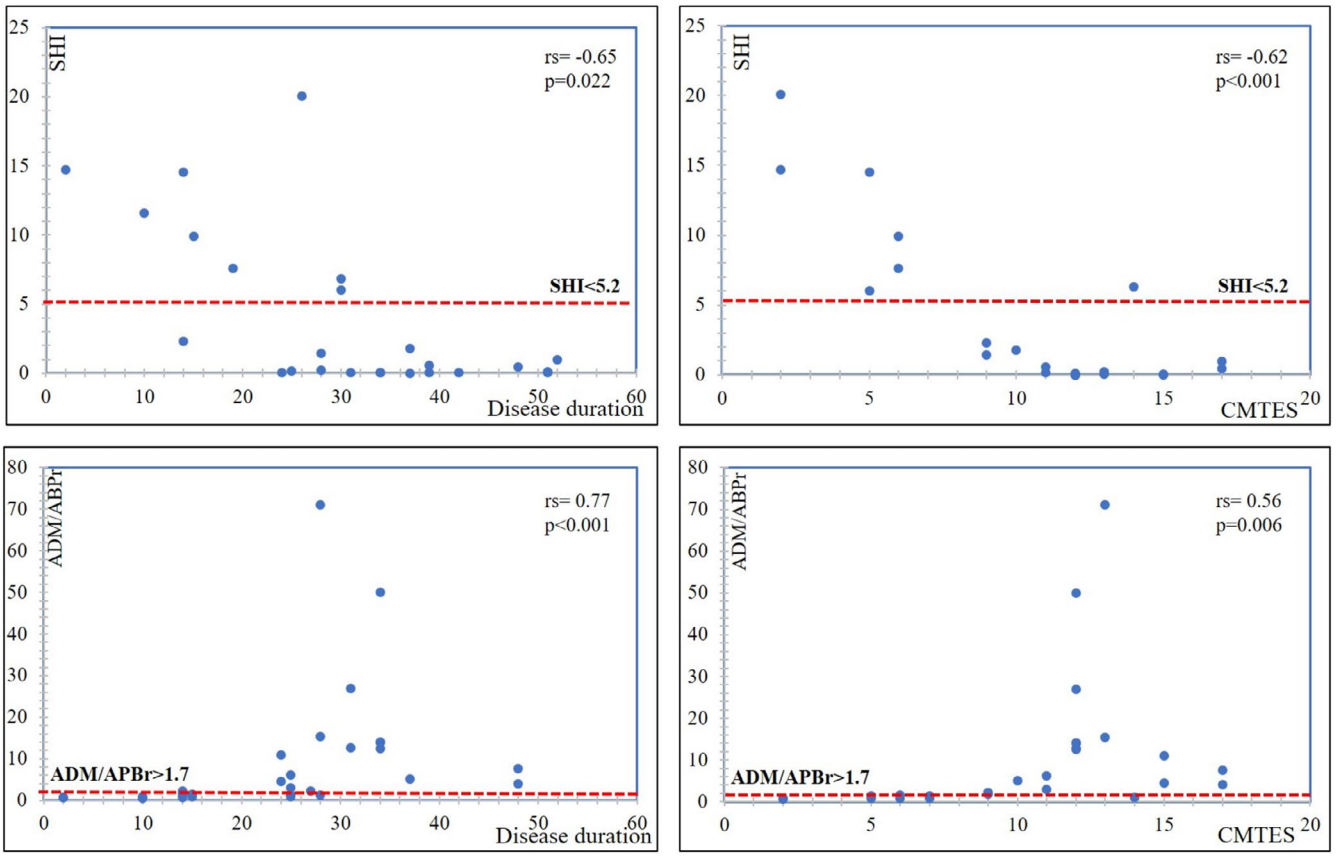

FIGURE 3 | Correlation between ADM/APBr and SHI with disease duration and severity. CMTES, CMT-Examination-Score; SHI, Split hand index; rs, Spearman's rho correlation. Red dotted lines identify ADM/APBr and SHI pathological cut-off of > 1.7 and < 5.2, respectively, which were deemed highly specific for ALS.

In our cohort, up to 59% of patients with CMTX1 presented with a disproportionate weakness and atrophy pattern in hands, which is not attributable to median/ulnar dissociation, as previously reported [11], but rather to the thenar/hypothenar differential involvement, as both FDI (ulnar) and APB (median) tended to be more involved than ADM (ulnar).

We assessed SHS at three different levels: 50% of patients had clinical SHS, 64% had neurophysiological SHS, and 38% had neuroradiological SHS. Taken together, clinical evaluation, neurophysiology, and neuroradiology indicate that up to 59% of CMTX1 in our whole cohort had SHS.

We found that the degree of neurophysiological SHS strongly correlated with disease duration and severity in CMTX1, indicating that, as the disease progressed, dissociated atrophy in hands became more evident. All CMTX1 males and females with disease duration > 15 and 28 years, respectively, showed neurophysiological SHS; however, this indicates that the process is much slower compared to ALS.

Conversely, SHS was not influenced by hand dominance and gender. Independence from gender might point towards an intrinsic liability of CMTX1 to preferential wasting in APB/FDI, since females with CMTX1 are usually much less affected as compared to males, due to the lyonization phenomenon [14, 15]. Moreover, concerning hand dominance independence, in our right-handed CMTX1 cohort, we were not able to find differences between the right (more liable to overuse) and left side in hand involvement, as SHS mostly presented bilaterally and was not more severe in the right, dominant side.

Although many previous studies on motor neuronopathies were focused on neurophysiology to define SHS, we are the first to perform hand muscle MRI to detect what we called the "radiological SHS". Indeed, to date, muscle MRI is considered the most reliable method to evaluate muscle fat infiltration and atrophy [18], especially in patients with normal nerve conduction studies. Furthermore, we were also able to detect active denervation, which precedes muscle atrophy, through STIR [19] sequences in APB/FDI but not in ADM, both in patients who were symptomatic (either clinically, or neurophysiologically, or radiologically) or even asymptomatic (two patients) for SHS (Table 3, Figure 4).

Functional consequences of SHS are relevant and involve mainly the ability to manipulate everyday tools. We previously found that 79% and 41% of males and 79% and 27% of females with CMTX1 in the Italian CMT Registry cohort had difficulties with buttons and cutlery, respectively, which was a much higher rate than other common CMT subtypes (e.g., 53% and 9%, respectively, in CMT1A; 46% and 16%, respectively, in CMT1B) [12]. This significantly impacts patients' quality of life.

Apart from CMTX1, reports of thenar-hypothenar SHS in CMT are limited to a few cases with *GARS*- [20], *SIGMAR1*- [21],

TABLE 3 | Radiological data of patients with CMTX1.

Patient number	Gender	MRC (APB/FDI/ADM) R	MRC (APB/FDI/ADM) L	Fat fraction (APB/FDI/ADM) R	Fat fraction (APB/FDI/ADM) L	Atrophy scale (APB/FDI/ADM) R	Atrophy scale (APB/FDI/ADM) L	STIR hyperintensity (APB/FDI/ADM) R	STIR hyperintensity (APB/FDI/ADM) L
9	M	4−/4+/4	4+/4+/4	NA/NA/NA	NA/NA/NA	0/0/0	0/0/0	Yes/No/No	No/No/No
16	M	0/1/3	0/1/3	NA/NA/NA	NA/NA/NA	3/4/4	3/4/3	No/No/No	No/No/No
18	M	4+/4+/5	4+/4+/5	NA/NA/NA	4%/5%/7%	0/0/0	0/0/0	No/No/No	Yes/No/Yes
1	F	4+/4/4+	4+/4/4	9%/6%/12%	8%/6%/9%	1/0/1	1/0/1	Nes/No/No	Yes/No/No
13	M	1/1/4	3/2/4	15%/60%/15%	**56%/72%/11%**	**3/3/1**	**3/3/1**	Yes/No/No	Yes/No/No
8	M	4+/4+/4+	4/4+/4	6%/9%/10%	8%/9%/10%	0/0/0	0/0/0	No/No/No	No/No/No
4	F	0/2/4−	0/2/4−	**86%/80%/18%**	**87%/75%/24%**	**3/3/2**	**3/3/2**	Yes/No/No	Yes/No/No
6	F	1/1/3	3/3/3	**27%/22%/11%**	15%/37%/33%	**2/2/1**	**2/2/1**	Yes/Yes/No	Yes/Yes/No

Note: Highlighted in bold radiological SHS (see Section 2).
Abbreviations: ADM, abductor digiti minimi; APB, abductor pollicis brevis; F, female; FDI, first dorsal interosseus; L, left; M, male; MRC, Medical Research Council (MRC) Scale; na, not available; R, right; STIR, Short-TI Inversion Recovery.

AARS- [22], and *BSCL2*-related [23, 24] neuropathy. Interestingly, variants in *GARS* [25] and *SIGMAR1* [26] are variably associated with both motor-predominant neuropathic/neuronopathic forms. Some cases with *AARS*-related myeloneuropathy [27] have been reported as well. Variants in *BSCL2* cause a wide spectrum of conditions ranging from hereditary spastic paraparesis to motor-predominant CMT2, neuronopathic/ALS-like forms [28]. Indeed, CMTX1 is arguably the only pure, sensorymotor, length-dependent polyneuropathy in which SHS has been consistently described to date.

Hence, we evaluated the specificity of ADM/APBr > 1.7 neurophysiological criterion, as a useful tool to discriminate CMTX1 from other CMT subtypes. The advantage of ADM/APBr compared to SHI is that the ulnar nerve is derived from ADM in most patients. Moreover, in our cohort, ADM/APBr strongly correlated with SHI (rs = −0.92). We found that ADM/APBr > 1.7 yielded a specificity of 81% for CMTX1 among all CMTs and was more specific for CMTX1 males among I-CMT/CMT1 (85%) as compared to females among CMT2/I-CMT (74%).

Pathogenesis of SHS is still unknown. Over the years, both central and peripheral theories, the former relying on upper/lower motor neuron dysfunction, the latter on motor peripheral axon/endplate changes, have been postulated [1, 2].

Our findings on thenar-hypothenar SHS in CMTX1 are important because they corroborate the key role of PNS in the genesis of SHS. Indeed, even though subclinical involvement of CNS is demonstrated by evoked potential abnormalities [29] and exceptional cases of reversible stroke-like episodes [30, 31] have been reported, CMTX1 neuropathology predominantly, if not exclusively, affects PNS at gap junctions in non-compact myelin (paranodal loops and Schmidt–Lanterman incisures) [32].

As far as peripheral motor axons are concerned, increased excitability in APB and/or FDI-innervating axons compared to ADM has been previously described. Shibuya and colleagues [33] demonstrated that, in ALS, motor neuronal death was accelerated by axonal excitability changes caused by increased nodal persistent sodium (and reduced potassium) current, which was especially observed in those axons supplying APB/FDI muscles compared to ADM. Similar findings of an increased excitability in APB were also detected in SBMA [9]. However, in 2009, Bae and colleagues [34] found that nodal persistent sodium conductances were physiologically more prominent in FDI axons compared to ADM in normal subjects as well.

In the only study on axon excitability in CMTX1 performed to date, Liang and colleagues [35] found a slightly increased nodal K^+ fast currents in APB muscle compared to non-CMTX1 controls, leading to modest hyperpolarization of the resting membrane potential. This modest hyperpolarization, the authors speculated, could contribute to a decrease in refractoriness and relative refractory period, eventually leading toward an increase in excitability.

As far as the motor-end plate, one study revealed that the percentage of area decrement on repetitive nerve stimulation was physiologically significantly greater in APB/FDI compared to ADM [36]. This indicates that physiological variability at the

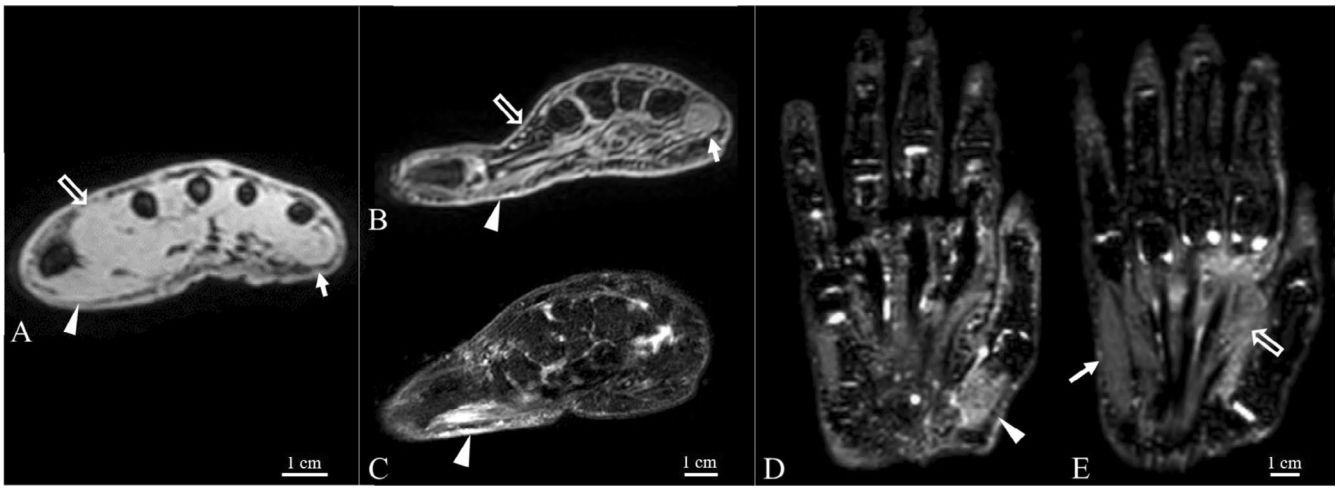

FIGURE 4 | Hand muscles MRI in CMTX1 patients. Normal APB, FDI, and ADM muscles signal and trophism in patient 8 (A, Axial Dixon T1w, water image). Severe atrophy of APB and FDI with relative sparing of ADM (B, Axial Dixon T1w, water image), and signs of active denervation in APB (C, axial STIR) in patient 13. STIR hyperintensity in APB (D) and FDI (E) but not ADM (E), reflecting dissociated active denervation, on the coronal plane in patient 6. APB – arrowhead; FDI – open arrow; ADM – arrow. ADM, abductor digiti minimi; APB, abductor pollicis brevis; FDI, first dorsal interosseous.

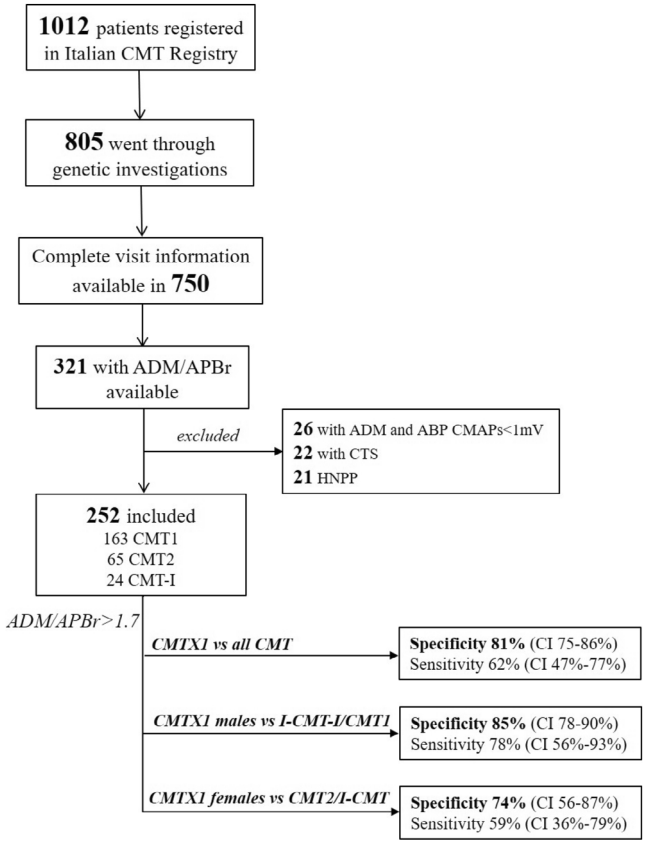

FIGURE 5 | Assessment of the specificity of SHS for CMTX1 among CMTs through ADM/APBr. ADM, abductor digiti minimi; APB, abductor pollicis brevis; CMAP, Compound muscle action potential; CTS, carpal tunnel syndrome; HNPP, Hereditary Neuropathy with Liability to Pressure Palsies.

neuromuscular junction in small hand muscles may determine intrinsic differential muscle vulnerability, potentially contributing to the development of the SHS.

We acknowledge a few limitations to this study. First, we excluded from the study patients with CTS based on clinical symptoms or previous history of surgery, rather than neurophysiological criteria. Second, as far as Aim 2 is concerned, we acknowledge the limitation of using retrospective data. Third, we were able to perform hand muscle MRI only in a few patients. Some patients living very distant from the centre refused to perform MRI; some others were incapable of remaining still with their hands during MRI due to hand weakness/tremor. As MRI is a novel approach to SHS investigation, only larger studies will help draw robust conclusions on its sensitivity and specificity.

In conclusion, we demonstrated that up to 59% of patients with CMTX1 develop SHS, which is an atrophy pattern in the hand characterized by thenar-hypothenar dissociated atrophy. In CMTX1, SHS worsens along with disease severity and duration, and is independent of gender and hand dominance. Interestingly, MRI STIR sequences might be useful to detect the dissociated thenar-hypothenar denervation pattern in patients still asymptomatic for SHS.

Moreover, ADM/APBr could potentially be a useful diagnostic tool to properly address genetic testing (rule in) for *GJB1*, especially in males. Concurrently, as variants of uncertain significance (VUS) are increasingly found with next-generation sequencing techniques, SHS, when present, might be helpful to support, with prudence, *GJB1* VUS pathogenicity in males with demyelinating/intermediate neuropathy.

Functional consequences of SHS are relevant. It is therefore crucial for clinicians to recognize when SHS is present and provide advice to improve patients' quality of life, such as using customized cutlery (thicker, with ergonomic handles or modified grips), Velcro fastenings instead of buttons, or adaptive keyboards or voice control devices for computers and phones. A multidisciplinary approach with an occupational therapist should always be pursued.

Pathogenesis of SHS remains extremely unclear, as SHS has been described over the years in ALS-upper motor neuron predominant forms, ALS-lower motor neuron predominant forms, pure lower motor neuronopathies, and, with CMTX1, peripheral sensorimotor neuropathy. Further studies on axonal excitability in CMTX1 are required to further entangle the pathophysiological role of PNS in the development of SHS.

Author Contributions

Alessandro Bertini: conceptualization, writing – original draft, methodology, writing – review and editing, formal analysis, data curation. **Marco Moscatelli:** investigation, methodology. **Claudia Ciano:** investigation. **Mattia Verri:** investigation. **Eleonora Cavalca:** investigation, project administration. **Luca Maria Sconfienza:** methodology. **Marina Grisoli:** investigation. **Paola Lanteri:** investigation. **Davide Pareyson:** writing – original draft, funding acquisition, writing – review and editing, investigation. **Chiara Pisciotta:** conceptualization, investigation, writing – original draft, writing – review and editing.

Acknowledgements

Alessandro Bertini, Eleonora Cavalca, Davide Pareyson, and Chiara Pisciotta are members of the Inherited Neuropathy Consortium RDCRN and the European Reference Network for Rare Neuromuscular Diseases (ERN EURO-NMD). Open access funding provided by BIBLIOSAN. Fiore Manganelli, Gian Maria Fabrizi, Angelo Schenone, Stefano Tozza, Tiziana Cavallaro, Federica Taioli, Moreno Ferrarini, Marina Grandis, Emilia Bellone, Paola Mandich, Stefano C. Previtali, Yuri Falzone, Isabella Allegri, Luca Padua, Costanza Pazzaglia, Andrea Quattrone, Paola Valentino, Luca Gentile, Massimo Russo, Isabella Moroni, Emanuela Pagliano, Paola Saveri, Stefania Magri, Franco Taroni, Anna Mazzeo, Lucio Santoro, Giuseppe Vita.

Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

Data available on request due to privacy/ethical restrictions.

References

1. P. Corcia, P. Bede, P. F. Pradat, P. Couratier, S. Vucic, and M. de Carvalho, "Split-Hand and Split-Limb Phenomena in Amyotrophic Lateral Sclerosis: Pathophysiology, Electrophysiology and Clinical Manifestations," *Journal of Neurology, Neurosurgery, and Psychiatry* 92 (2021): 1126–1130.

2. A. Eisen and S. Kuwabara, "The Split Hand Syndrome in Amyotrophic Lateral Sclerosis," *Journal of Neurology, Neurosurgery, and Psychiatry* 83 (2012): 399–403.

3. T. Chiba, F. Konoeda, M. Higashihara, et al., "C8 and T1 Innervation of Forearm Muscles," *Clinical Neurophysiology* 126 (2015): 837–842.

4. A. J. Wilbourn, "The 'Split Hand Syndrome'," *Muscle & Nerve* 23 (2000): 138.

5. S. Kuwabara, M. Sonoo, T. Komori, et al., "Dissociated Small Hand Muscle Atrophy in Amyotrophic Lateral Sclerosis: Frequency, Extent, and Specificity," *Muscle & Nerve* 37 (2008): 426–430.

6. J. Fang, M.-S. Liu, Y.-Z. Guan, et al., "Pattern Differences of Small Hand Muscle Atrophy in Amyotrophic Lateral Sclerosis and Mimic Disorders," *Chinese Medical Journal* 129 (2016): 792–798.

7. J. Kalita, S. Kumar, U. K. Misra, and Z. Neyaz, "Split Hand Index and Ulnar to Median Ratio in Hirayama Disease and Amyotrophic Lateral Sclerosis," *Amyotroph Lateral Scler Frontotemporal Degener* 18 (2017): 598–603.

8. P. Menon, M. C. Kiernan, C. Yiannikas, J. Stroud, and S. Vucic, "Split-Hand Index for the Diagnosis of Amyotrophic Lateral Sclerosis," *Clinical Neurophysiology* 124 (2013): 410–416.

9. K. Shibuya, S. Misawa, A. Uzawa, et al., "Split Hand and Motor Axonal Hyperexcitability in Spinal and Bulbar Muscular Atrophy," *Journal of Neurology, Neurosurgery, and Psychiatry* 91 (2020): 1189–1194.

10. A. Abraham, Y. Fainmesser, V. E. Drory, and V. Bril, "Split-Hand Phenomenon in Motor Neuron Diseases: Sonographic Assesment of Muscle Thickness," *Clinical Neurophysiology* 131 (2020): 1721–1725.

11. P. J. Tomaselli, A. M. Rossor, A. Horga, et al., "Mutations in Noncoding Regions of GJB1 Are a Major Cause of X-Linked CMT," *Neurology* 88 (2017): 1445–1453.

12. C. Pisciotta, A. Bertini, I. Tramacere, et al., "Clinical Spectrum and Frequency of Charcot-Marie-Tooth Disease in Italy: Data From the National CMT Registry," *European Journal of Neurology* 30 (2023): 2461–2470.

13. S. M. Murphy, D. N. Herrmann, M. P. McDermott, et al., "Reliability of the CMT Neuropathy Score (Second Version) in Charcot-Marie-Tooth Disease," *Journal of the Peripheral Nervous System* 16 (2011): 191–198.

14. S. M. Murphy, R. Ovens, J. Polke, et al., "X Inactivation in Females With X-Linked Charcot-Marie-Tooth Disease," *Neuromuscular Disorders* 22 (2012): 617–621.

15. C. E. Siskind, S. M. Murphy, R. Ovens, J. Polke, M. M. Reilly, and M. E. Shy, "Phenotype Expression in Women With CMT1X," *Journal of the Peripheral Nervous System* 16 (2011): 102–107.

16. C. J. Record, M. Skorupinska, M. Laura, et al., "Genetic Analysis and Natural History of Charcot-Marie-Tooth Disease CMTX1 due to GJB1 Variants," *Brain* 146 (2023): 4336–4349.

17. F. B. Panosyan, M. Laura, A. M. Rossor, et al., "Cross-Sectional Analysis of a Large Cohort With X-Linked Charcot-Marie-Tooth Disease (CMTX1)," *Neurology* 89 (2017): 927–935.

18. C. M. Doherty, J. M. Morrow, R. Zuccarino, et al., "Lower Limb Muscle MRI Fat Fraction Is a Responsive Outcome Measure in CMT X1, 1B and 2A," *Annals of Clinical Translational Neurology* 11 (2024): 607–617.

19. C. M. McDonald, G. T. Carter, R. C. Fritz, M. W. Anderson, R. T. Abresch, and D. D. Kilmer, "Magnetic Resonance Imaging of Denervated Muscle: Comparison to Electromyography," *Muscle & Nerve* 23 (2000): 1431–1434.

20. J. Jiménez-Jiménez, I. Navarrete, I. Azorín, et al., "Insights Into Phenotypic Variability Caused by GARS1 Pathogenic Variants," *European Journal of Neurology* 31 (2024): e16416.

21. S. C. Previtali, E. Zhao, D. Lazarevic, et al., "Expanding the Spectrum of Genes Responsible for Hereditary Motor Neuropathies," *Journal of Neurology, Neurosurgery, and Psychiatry* 90 (2019): 1171–1179.

22. B. Bansagi, T. Antoniadi, S. Burton-Jones, et al., "Genotype/Phenotype Correlations in AARS-Related Neuropathy in a Cohort of Patients From the United Kingdom and Ireland," *Journal of Neurology* 262 (2015): 1899–1908.

23. B. P. Van de Warrenburg, H. Scheffer, J. J. van Eijk, et al., "BSCL2 Mutations in Two Dutch Families With Overlapping Silver Syndrome-Distal Hereditary Motor Neuropathy," *Neuromuscular Disorders* 16 (2006): 122–125.

24. M. Auer-Grumbach, W. N. Löscher, K. Wagner, et al., "Phenotypic and Genotypic Heterogeneity in Hereditary Motor Neuronopathy Type V: A Clinical, Electrophysiological and Genetic Study," *Brain* 123 (2000): 1612–1623.

25. R. Markovitz, R. Ghosh, M. E. Kuo, et al., "GARS-Related Disease in Infantile Spinal Muscular Atrophy: Implications for Diagnosis and

Treatment," *American Journal of Medical Genetics. Part A* 182 (2020): 1167–1176.

26. A. Al-Saif, F. Al-Mohanna, and S. Bohlega, "A Mutation in Sigma-1 Receptor Causes Juvenile Amyotrophic Lateral Sclerosis," *Annals of Neurology* 70 (2011): 913–919.

27. W. W. Motley, L. B. Griffin, I. Mademan, et al., "A Novel AARS Mutation in a Family With Dominant Myeloneuropathy," *Neurology* 84 (2015): 2040–2047.

28. A. A. Zambon, V. Pini, L. Bosco, et al., "Early Onset Hereditary Neuronopathies: An Update on Non-5q Motor Neuron Diseases," *Brain* 146 (2023): 806–822.

29. Q. Wen, L. Cao, and C. Yang, "The Electrophysiological Features in X-Linked Charcot-Marie-Tooth Disease With Transient Central Nervous System Deficits," *Frontiers in Neurology* 9 (2018): 461.

30. A. Sagnelli, G. Piscosquito, L. Chiapparini, et al., "X-Linked Charcot-Marie-Tooth Type 1: Stroke-Like Presentation of a Novel GJB1 Mutation," *Journal of the Peripheral Nervous System* 19 (2014): 183–186.

31. D. Tian, Y. Zhao, R. Zhu, Q. Li, and X. Liu, "Systematic Review of CMTX1 Patients With Episodic Neurological Dysfunction," *Annals of Clinical Translational Neurology* 8 (2021): 213–223.

32. L. J. Bone, S. M. Deschênes, R. J. Balice-Gordon, K. H. Fischbeck, and S. S. Scherer, "Connexin32 and X-Linked Charcot-Marie-Tooth Disease," *Neurobiology of Disease* 4 (1997): 221–230.

33. K. Shibuya, S. Misawa, S. Nasu, et al., "Split Hand Syndrome in Amyotrophic Lateral Sclerosis: Different Excitability Changes in the Thenar and Hypothenar Motor Axons," *Journal of Neurology, Neurosurgery, and Psychiatry* 84 (2013): 969–972.

34. J. S. Bae, S. Sawai, S. Misawa, K. Kanai, S. Isose, and S. Kuwabara, "Differences in Excitability Properties of FDI and ADM Motor Axons," *Muscle & Nerve* 39, no. 3 (2009): 350–354.

35. C. Liang, J. Howells, M. Kennerson, G. A. Nicholson, D. Burke, and K. Ng, "Axonal Excitability in X-Linked Dominant Charcot Marie Tooth Disease," *Clinical Neurophysiology* 125 (2014): 1261–1269.

36. M. De Carvalho and M. Swash, "The Split Hand in Amyotrophic Lateral Sclerosis: A Possible Role for the Neuromuscular Junction," *Amyotroph Lateral Scler Frontotemporal Degener* 20 (2019): 368–375.

Supporting Information

Additional supporting information can be found online in the Supporting Information section.