

IS THE TRIPLE STIMULATION TECHNIQUE A BETTER QUANTIFICATION TOOL OF MOTOR DYSFUNCTION THAN MOTOR EVOKED POTENTIALS IN MULTIPLE SCLEROSIS?

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Abstract

The triple stimulation technique (TST) was rarely used in multiple sclerosis (MS). This study aimed to compare TST and motor evoked potentials (MEP) for the quantification of motor dysfunction. Central motor conduction based on MEP (four limbs) and TST (upper limbs) was assessed in 28 MS patients with a median Expanded Disability Status Scale (EDSS) of 4. EDSS, timed 25-foot walk (T25FW), grasping strength and motor components of the MS functional composite were evaluated. Regression analysis was used to assess the relationship between MEP, TST and clinical findings. TST was negatively correlated with EDSS ($r = -0.74$, $p < 0.0001$) and to a lesser extent with T25FW ($r = -0.47$, $p < 0.05$), and grasping strength ($r = -0.43$, $p < 0.05$). A multiple regression analysis underlined the better correlation between clinical data and TST ($R^2 = 0.56$, $p < 0.0005$) than with MEP ($0.03 < R^2 < 0.22$, $p > 0.05$). This study evidenced the value of TST as a quantification tool of motor dysfunction. TST appeared to reflect a global disability since it was correlated not only to hand function but also to walking capacity.

Keywords Triple stimulation technique · Motor evoked potentials · Multiple sclerosis · Conduction failure · Axonopathy · Disability

Introduction

Transcranial magnetic stimulation (TMS) is a neurophysiological technique eliciting motor evoked potentials (MEP) useful for the diagnostic and functional assessment of the pyramidal track impairment [1, 2]. The diagnostic efficacy of MEP in multiple sclerosis (MS), however, is limited by the high sensitivity of magnetic resonance imaging (MRI) mainly at the brain level [3]. Conversely, to document pathological changes occurring during the course of MS, MRI is less useful because of its weak association with clinical findings, known as the “functional–anatomical paradox” [4], while MEP evaluating long eloquent motor pathway is better correlated with functional involvement [5–9].

The increased central motor conduction time (CMCT), theoretically related to the demyelination process, is the most widely used MEP parameter in clinical studies [8, 10, 11] despite weak correlations with the functional capacities [12, 13]. The MEP size reduction might be regarded as the axonal loss consequence [14] which is closely linked to disability, as demonstrated by MRI [15]. However, the motor response size is further influenced by the desynchronization of the TMS-induced motoneurones discharges, which makes the amplitude interpretation quite tricky [16].

The triple stimulation technique (TST), developed by Magistris [17], allows a precise quantification of the motor central conduction failures by the resynchronization of the upper motoneurone discharges. The TST has already been used to diagnose and monitor the functional consequences of several peripheral or central nervous conditions such as amyotrophic lateral sclerosis [18], Kennedy disease [19], spinocerebellar ataxia [20], multiple systemic atrophy [21], chronic inflammatory demyelinating polyradiculoneuropathy [22] and Guillain–Barre syndrome [23].

The test–retest repeatability of TST measurements in healthy subjects [24] and MS patients [25] showed that TST was particularly suited for follow-up examinations. The TST method was also used to study motor fatigue [26, 27]. In healthy subjects, during a fatiguing exercise, there was a TST amplitude ratio decrease, reflecting a decline in central motor conduction, which was more pronounced than in MS patients, while the reduction of force was similar [27]. Humm et al. used TST to quantify temperature-induced changes (Uhthoff phenomenon) in the pyramidal tract of MS patients, responsible for paroxysmic symptoms [28]. They observed a significant correlation between changes in walking velocity (after a cold bath) and the TST amplitude ratio. The temperature-related changes of central conduction block, as measured by TST, were present only for the patients exhibiting an abnormal prolongation of CMCT. Somehow, this is a proof of concept that TST could be a suitable tool in order to monitor patients concerned by the Uhthoff phenomenon and that CMCT could identify those who are more at risk to be exposed to temperature-induced conduction blocks. Firmin et al. found that in MS patients the contribution of slowly conducting portions of the central motor pathway was more important than in healthy subjects [29].

Studies using TST for the assessment of motor capacities in MS are scarce. The TST amplitude ratio was reduced in presence of pyramidal signs [12, 30], but was not significantly different in patients with

equivalent clinical motor deficit regardless their phenotype [12]. These authors also observed a marked increase in TST amplitude ratio after 5 days of intravenous methylprednisolone in patients with relapsing–remitting (RR) and secondary progressive (SP) MS, paralleling an increase in muscle force, whereas no increase in TST amplitude ratio was depicted for primary progressive (PP) patients [13]. Never addressed before, the purpose of this study was to compare TST and classical MEP parameters (latency and amplitude) for the functional quantification of upper and lower limb motor dysfunction.

Materials and methods

STUDY PATIENTS

This was a prospective transversal study conducted on a sample of 28 MS patients (16 women, 12 men) referred to the neurological department of the University Hospital of Liège (Belgium) between 2013 and 2015. The inclusion criteria were: (1) a diagnosis of MS based on the 2010 revised McDonald criteria [31], and (2) a clinically stable phase at the time of evaluation ensured by a minimum 3-month period between the previous relapse and the evaluation. Patients with a history of seizures or with implanted devices were excluded from the study. The Local Ethics Committee approved the study.

CLINICAL SCALE AND MEASUREMENTS

An MS specialized neurologist assessed the Expanded Disability Status Score (EDSS) and a trained technician performed the clinimetric tests. The clinimetry included the “9-Hole Peg Test (9HPT)” and the “Timed 25-Foot Walk (T25FW)”, the motor components of the Multiple Sclerosis Functional Composite scale (MSFCm) [32]. The 9HPT and T25FW assessment protocol was described in the MSFC Administration and Scoring Manual [32]. The Jamar dynamometer was used to measure the grasping strength (JAMAR) [33]. Z-scores were derived for each individual dimension (Z-9HPT, Z-T25FW, Z-JAMAR) based on a local presumably healthy population ($n = 60$). For Z-9HPT (adjusted for age) and Z-JAMAR (adjusted for age, height and sex), the average of both upper limbs was calculated. The Z-T25FW was adjusted for age and size. The clinical assessment took about 15 min.

MEP PROCEDURE

A six-channel Keypoint G4 apparatus (Natus Medical Incorporated) was used for the recordings and a Magstim 200 device (The Magstim Company Ltd., Whitland, UK) with a circular (70 mm inner diameter) hand-held coil (maximal output 2.2 T) for TMS. MEP values were recorded bilaterally from the *first dorsal interosseous* (FDI) muscle in the upper limbs (belly–tendon technique) and from the tibialis anterior (TA) muscle in the lower limbs (belly–tibial tubercle technique). Recording electrodes consisted of pre-gelled disposable surface electrodes (Alpine Biomed, REF 9013P0873). Corresponding ground electrode was taped to the forearm. The bandpass filter setting was set from 0.5 Hz to 2 kHz

for upper limbs and from 0.5 Hz to 3 kHz for lower limbs. The motor threshold (minimum intensity required to evoke a discernible motor response in 50% of trials at an amplification of 100 μ V per division) was first measured with muscle at rest. Slight coil displacements were made until the lowest threshold was found. TMS was performed at a 130% above motor threshold intensity (the best MEP recording was selected out of 5 trials). Cervical (C8) and lumbar (L5) foraminal electromagnetic stimulation was delivered at 80% and 100% of maximal output (2.2 T), respectively. A slight isometric voluntary contraction (approximately 20% of the maximal isometric voluntary contraction) was asked during TMS only.

For each limb, two parameters were considered: (1) the central motor conduction time (CMCT), obtained by subtracting the peripheral conduction time elicited by magnetic stimulation of the cervical and lumbar spinal roots from the TMS-induced response latency; and (2) the amplitude ratio (AR) defined as the ratio between the baseline-to-peak amplitude of the TMS-induced response divided by the baseline-to-peak amplitude of the compound muscle action potentials (CMAP) obtained by a supramaximal electric stimulation applied distally to the peripheral nerve (ulnar nerve at the wrist and common peroneal nerve at the head of fibula). CMAP were evoked by 0.2 ms constant current square waves through bipolar surface stimulation with two 7-mm-diameter felt tip pads, 2.3 cm apart (Natus Medical Incorporated, REF 9013L0362).

TST PRINCIPLE

The TST protocol has been described in detail [17, 30]. Because of the invasive procedure for the lower limb investigation (needle stimulation of the sciatic nerve at the level of gluteal fold), the recordings were restricted to upper limbs [34]. The equipment used was the same as for MEP measurements with a specific commercial TST software (Natus Medical Incorporated). During the examinations, patients lay comfortably in an armchair with the palms turned up. The TST principle is briefly outlined hereafter.

For resynchronizing central motor volley, three stimuli were applied successively with specific delays leading to peripheral collisions. A supramaximal TMS (brain stimulation at 130% above motor threshold obtained at rest) was first applied depolarizing all recruitable cortical motoneurons supplying the target *abductor digiti minimi* (ADM) muscle (belly–tendon technique). After a first delay, a second stimulus (supramaximal electric stimulation) was applied to the ulnar nerve at wrist (distal peripheral nerve stimulation with bipolar electrode), resulting in a collision between the ascending action potentials from the wrist and the descending discharges from the cortex. Consequently, only the second stimulation (at wrist) induced a motor response recorded from the ADM muscle. Concomitant distal peripheral stimulation of ulnar and median nerves has been proposed by Humm et al. in order to take into account the “volume conduction phenomenon” [24, 35]. This technical adaptation is important when thenar muscles are recorded, because of the mixed (ulnar and median) innervation of this part of the hand. However, conduction volume is less likely to occur for the hypothenar muscles due to anatomical reason [17]. Thus, we have decided not to stimulate simultaneously ulnar and median nerve for this TST protocol targeting ADM. After a short time period (second delay), a third stimulus (supramaximal electric stimulation) was applied to the Erb’s point

(proximal peripheral nerve stimulation with monopolar electrode), resulting in a second motor response recorded from the ADM muscle. Delays were tailored to each patient, prior the actual TST recording. The first delay, between brain and distal peripheral stimulations, was defined as the difference between minimal MEP latency (rounded down to the nearest ms) and CMAP distal latency (rounded up to the nearest ms). The second delay, between proximal and distal peripheral stimulations, was defined as the difference between CMAP proximal latency (rounded down to the nearest ms) and CMAP distal latency (rounded up to the nearest ms). In case of central axonal failure, the collision took place between wrist and Erb stimulations, resulting in a TST response reduced in proportion of cortical neurones which were not brought to discharge by the TMS. For the central conduction failure quantification, the size of the first negative deflection of the TST test (TST_{test}, described above) was expressed as a percentage of the size of the first negative deflection of the TST control (TST_{ctl}). The latter was recorded by replacing the TMS by a supramaximal electric stimulation to the Erb's point with appropriate adjustment of the first delay (identical to the second delay) (Fig. 1). The TST_{test}/TST_{ctl} ratio reflects the proportion of the spinal motoneurons activated. To limit the discomfort related to the procedure, no more than five trials of TST were recorded in each patient. The TST amplitude ratio closest to 100% was selected. Each TST session lasted about 60 min.

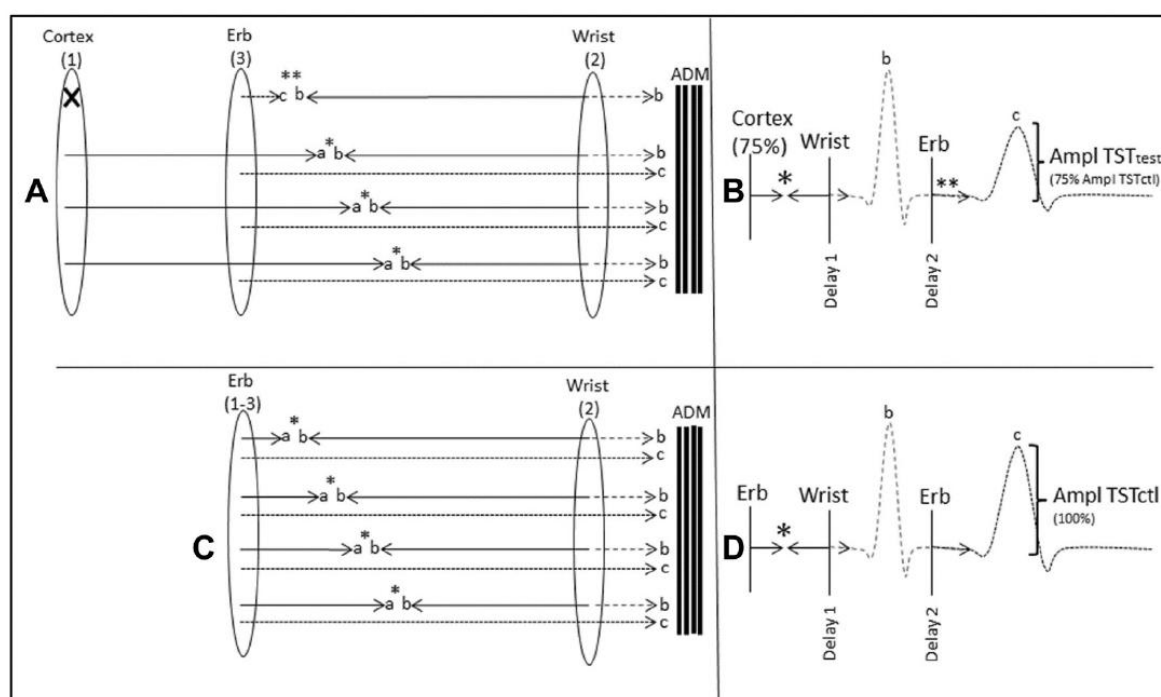


Fig. 1 Illustration of the triple stimulation technique (TST). **a, b** (TST_{test}) In this example, three out of four motoneurons are brought to discharge by a supramaximal TMS (1). After a delay, a supramaximal electric stimulation to the ulnar nerve at wrist (2) induces a first response from the *abductor digiti minimi* (ADM) muscle (b). Due to the collision between descending and ascending discharges (a*b), no response is elicited by the TMS. A third supramaximal electric stimulation applied at Erb's point (3), induces a second response recorded from

the ADM muscle (c). In case of central conduction failure, the collision occurs between descending (from Erb's point) and ascending (from ulnar nerve stimulation) discharges (c**b). The size reduction of the second response is due to the proportion of conduction failures. **c, d (TSTctI)**: The TSTtest is compared to a TSTctI (TMS replaced by Erb stimulation). Only one type of collision occurs, between descending (from first Erb stimulation) and ascending (from ulnar nerve stimulation) discharges (a*b). Thus the third stimulation, applied at Erb's point, induces a second ADM muscle response that is maximal (100%). The amplitude ratio between the second response of the TSTtest and the TSTctI provides the central conduction failure.

DATA ANALYSIS

Results were generally summarized by median and interquartile range (IQR) for quantitative data and by frequency tables for categorical data. The Pearson correlation coefficient (r) was used to measure the association between any two quantitative variables. Multiple regression analysis was used to assess the association between electrophysiology (dependent variable) and clinical data (predictors) in MEP and TST. The adjusted multiple coefficient of determination (R^2) was used to measure the quality of the relationship. Results were considered significant at the 5% critical level ($p < 0.05$). Statistical calculations were performed with the R software (version 3.4.3).

Results

Clinical and electrophysiological characteristics of study patients are displayed in Table 1. The median age was 47.2 years and the disease duration 16.2 years. The median EDSS was 4 (IQR 3.8–5.8). Upper limb AR (median 12.9%, IQR 7.3–22.9%) and lower limb AR (13.3%, IQR 7.9–22.2%) were more reduced compared to median TSTtest/TSTcontrol ratio (62.6%, IQR 35.9–85.8%) but fell just below the lower AR reference limit. The median difference between TST and upper limb AR was 41.7% (IQR 27.8–64.3%).

Table 1 Demographic, clinical and electrophysiological characteristics of MS study patients

Variable	N	Median (IQR)	Range	Norm ^a
RR/SP/PP	18/8/2			
Gender M/F	12/16			
Age (years)	28	47.2 (38.6–57.8)	21.0 to 73.0	
Disease duration (years)	28	16.2 (13.0–21.8)	7.5 to 36.5	
EDSS	28	4.0 (3.8–5.8)	2.0 to 8.5	
Z-JAMAR	28	1.1 (0.6–2.2)	–0.4 to 4.7	2.0
Z-9HPT	28	0.8 (0.2–3.6)	–1.7 to 51.2	2.0
Z-T25FW	28	4.6 (2.5–15.1)	0.4 to 327.2	2.0
AR UL (%)	28	12.9 (7.3–22.9)	3.7 to 56.5	14.6
CMCT UL (ms)	28	9.4 (8.2–13.0)	5.7 to 25.1	8.1
AR LL (%)	28	13.3 (7.9–22.2)	4.0 to 67.1	14.7
CMCT LL (ms)	28	23.8 (19.7–33.7)	16.5 to 53.4	16.6
TST (%)	28	62.6 (35.9–85.8)	9.8 to 97.6	93.0 ^b
TST–AR UL (%) ^c	28	41.7 (27.8–64.3)	–2.01 to 89.7	30.0 ^b

IQR interquartile range (P25–P75), *RR* relapsing/remitting, *SP* secondary progressive, *PP* primary progressive, *M* male, *F* female, *EDSS* Expanded Disability Status Scale, *Z* z-score, *JAMAR* grasping strength, *9HPT* Nine-Hole Peg Test, *T25FW* Timed 25-Foot Walk, *AR* amplitude ratio, *CMCT* central motor conduction time, *UL* upper limb, *LL* lower limb, *TST* triple stimulation test

^aNormative values were derived from a sample of presumably healthy subjects ($n=55$) in our laboratory unless indicated

^bNorms derived from triple stimulation technique [17]

^cDifference between TST and AR UL as an estimation of the amount of central desynchronization

Correlations between electrophysiological parameters are displayed in Table 2. Significant negative correlations were observed between CMCT and AR in the left upper limb ($r = -0.53$; $p < 0.001$) and in the right lower limb ($r = -0.46$; $p < 0.05$). CMCT measurements at the upper limb level and at the lower limb level were strongly correlated with each other ($p < 0.0001$) at both the right ($r = 0.62$) and left ($r = 0.66$) body sides. This was also the case for the correlations between upper and lower limbs AR ($r = 0.55$ and $r = 0.67$ for the right and left body sides, respectively; $p < 0.005$). TST was mainly correlated to AR, even though the higher correlation (with the left upper limb AR) did not exceed 0.63 ($p < 0.0001$).

Table 2 Pearson correlation coefficients between MEP parameters (CMCT, AR) and TST ($TST_{test}/TST_{control}$) recorded in lower and upper limbs of right and left body sides ($n=28$)

	CMCT r-UL	AR r-UL	CMCT l-UL	AR l-UL	CMCT r-LL	AR r-LL	CMCT l-LL	AR l-LL	TST r-UL	TST l-UL
CMCT r-UL	1.00									
AR r-UL	-0.35	1.00								
CMCT l-UL	0.74***	-0.39*	1.00							
AR l-UL	-0.34	0.78***	-0.53**	1.00						
CMCT r-LL	0.62***	-0.47*	0.56**	-0.40*	1.00					
AR r-LL	-0.31	0.55**	-0.24	0.59**	-0.46*	1.00				
CMCT l-LL	0.42*	-0.19	0.66***	-0.37	0.41*	-0.13	1.00			
AR l-LL	-0.13	0.38*	-0.24	0.67***	-0.11	0.44*	-0.35	1.00		
TST r-UL	-0.23	0.44*	-0.16	0.40*	-0.45*	0.49*	-0.02	0.16	1.00	
TST l-UL	-0.18	0.31	-0.39*	0.63***	-0.30	0.25	-0.45*	0.49*	0.53**	1.00

MEP motor evoked potentials, CMCT central motor conduction time, AR amplitude ratio, TST (triple stimulation technique) TST_{test}/TST_{ctl} ratio, r right body side, l left body side, UL upper limb, LL lower limb

* $p < 0.05$, ** $p < 0.005$, *** $p < 0.0005$

TST correlated significantly with the EDSS and clinimetry, except for 9HPT. It is worth emphasizing the high negative correlation between TST and EDSS ($r = -0.74$; $p < 0.0001$) (Table 3; Fig. 2) and, to a lesser extent, T25FW ($r = -0.47$; $p < 0.05$) and JAMAR ($r = -0.43$; $p < 0.05$) (Table 3). More than 50% of the EDSS variability was predicted by TST, the lower the TST, the higher the EDSS (Fig. 2). Correlations between MEP parameters and EDSS were lower but still significant ($0.40 < r < 0.54$; $p < 0.05$). A significant but weak correlation was observed between CMCT recorded in the lower limb and, respectively, 9HPT ($r = 0.39$; $p < 0.05$) and T25FW ($r = 0.44$; $p < 0.05$) (Table 3).

Table 3 Pearson correlation coefficients between electrophysiological and clinical data ($n=28$)

	CMCT UL	AR UL	CMCT LL	AR LL	TST UL
EDSS	0.40*	-0.47*	0.54*	-0.54*	-0.74***
Z-Jamar	0.16	-0.32	0.32	-0.13	-0.43*
Z-9HPT	0.30	-0.33	0.39*	-0.21	-0.31
Z-T25FW	0.27	-0.25	0.44*	-0.30	-0.47*

CMCT central motor conduction time, AR amplitude ratio, TST (triple stimulation technique) TST_{test}/TST_{ctl} ratio, UL upper limb, LL lower limb, EDSS Expanded Disability Status Scale, Z z-score, JAMAR grasping strength, 9HPT Nine-Hole Peg Test, T25FW Timed 25-Foot Walk

* $p < 0.05$, ** $p < 0.001$, *** $p < 0.0001$

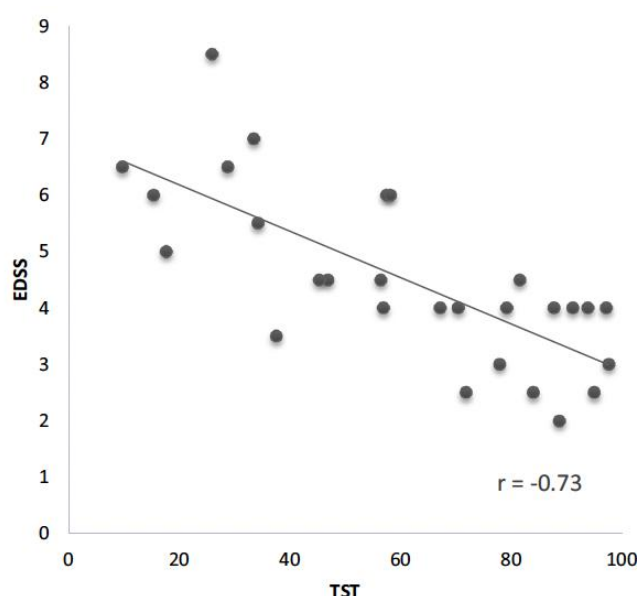


Fig. 2 Relationship between EDSS and conduction failure quantified by the triple stimulation technique (TST). *EDSS* Expanded Disability Status Scale, *TST* TST_{test}/TST_{ctl} ratio

Multiple regression analysis applied to TST (dependent variable) and clinical parameters (EDSS, 9HPT, T25FW and JAMAR as independent factors) evidenced a highly significant and stronger association (adjusted $R^2 = 0.56$; $p < 0.0005$) than between each MEP component and the same clinical parameters (adjusted R^2 ranging from 0.03 to 0.22 with p value between 0.045 and 0.34) (Table 4).

Table 4 Multiple regression analysis between electrophysiology (dependent variable) and clinical parameters (predictors) in MS study patients ($n=28$)

Predictor	Regression coefficient $\pm$ SE				
	Model 1—TST $R^2=0.56$ $p<0.0005$	Model 2—CMCT UL $R^2=0.03$ $p=0.34$	Model 3—CMCT LL $R^2=0.19$ $p=0.061$	Model 4—AR UL $R^2=0.13$ $p=0.13$	Model 5—AR LL $R^2=0.22$ $p=0.045$
Intercept	134.8 ± 14.7	5.87 ± 3.56	12.8 ± 8.18	0.40 ± 0.10	0.43 ± 0.09
EDSS	-15.6 ± 3.60	1.16 ± 0.87	2.98 ± 2.00	-0.05 ± 0.03	-0.06 ± 0.02
9HPT	0.75 ± 0.41	0.05 ± 0.10	0.03 ± 0.23	-0.01 ± 0.01	0.01 ± 0.01
T25FW	0.02 ± 0.06	-0.01 ± 0.01	0.01 ± 0.03	0.01 ± 0.01	0.01 ± 0.01
JAMAR	-6.25 ± 3.50	-0.23 ± 0.85	0.98 ± 1.96	-0.02 ± 0.02	0.01 ± 0.02

TST (triple stimulation technique) TST_{test}/TST_{ctl} ratio, *CMCT* central motor conduction time, *AR* amplitude ratio, *UL* upper limb, *LL* lower limb, *EDSS* Expanded Disability Status Scale, *9HPT* Nine-Hole Peg Test, *T25FW* Timed 25-Foot Walk, *JAMAR* grasping strength, R^2 adjusted multiple coefficient of determination, *SE* standard error

Discussion

Transcranial magnetic stimulation is widely used both for research and clinical monitoring in MS. In agreement with the literature [9, 36–38], the value of classical MEP parameters (CMCT, AR) as quantitative tools for the functional motor assessment was confirmed by the good correlations

observed with the EDSS (Table 3). Moreover, most of the electrophysiological parameters were correlated with each other. The highly significant correlations observed between the upper limb CMCT/AR and the lower limb CMCT/AR for the right and left body sides could be explained by a strong influence of the cervical MS lesions on both nervous conduction to arms and legs (Table 2).

Studies addressing the relation between electrophysiology and clinimetry in MS are scarce. Kalkers et al. identified a significant correlation between CMCT and T25FW but no association was found with the 9HPT [11]. However, in another study [39], CMCT was correlated with hand dexterity and phasic strength. Our results, characterized by a significant correlation between MSFC (9HPT and T25FW) and CMCT in the lower limbs, comfort the ability of MEP to document the four limbs motor function (Table 3).

To date, TST has not been used frequently in the field of MS. This novel work evidenced a good correlation between clinical and TST data (Table 3). Actually, the association was notably better than with the classical MEP parameters as revealed by multiple regression analysis (Table 4). As TST findings, restricted to upper limbs, were not only correlated to hand function (grasping strength) but also to walking speed (T25FW, Table 3), we believe that TST can be a suitable electrophysiological marker of disability and functional impairment involving the four limbs and that TST should be used to complement classical MEP, especially CMCT in the lower limbs. The weak association between TST and hand dexterity could be explained by a more sophisticated task required by the 9HPT, involving not only muscle strength but also somatosensory afferences, coordination as well as visual function.

MRI data demonstrate a close link between axonopathy and irreversible functional deficit [15, 40]. The TST amplitude ratio reduction cannot distinguish axonal loss from conduction block [30]. However at a chronic stage (i.e., a permanent or long lasting conduction block), both neurophysiopathological processes should induce an equivalent clinical expression. The interpretation of the alteration of MEP amplitude is less obvious given the variety of mechanisms potentially involved. In the MS population, MEP AR is usually more affected and variable than TST [30]. Pathological substrates for these differences between both techniques could include an inhomogeneous conduction velocity among the pyramidal fibers, together with a reduced firing rate resulting in a lower temporo-spatial summation at synaptic level in the spinal cord [14]. In our MS population, TST values were much higher than AR evaluated by MEP (median 62.6% vs. 12.9%, Table 1). Probably linked to the high inter-individual variability of the desynchronization, there was also a relatively weak correlation between TST and MEP (Table 2). Moreover, the difference between TST and upper limb AR (median 41.7%), which estimates the amount of central desynchronization, suggested, in agreement with a previous study based on the *peristimulus time histograms* technique [41], an increased temporal dispersion of the central descending volleys in MS compared to healthy subjects (estimated at 30%) [17]. Consequently, MEP AR cannot be used for the extrapolation of axonopathy. By resynchronization of central motor volley, which was objective in the current work (Table 1), TST is theoretically a better clue of central motor axonopathy. As the latter is linked to irreversible functional deficit [15, 40], the close association ($R^2 = 0.56$) between TST and clinical data, assessed by multiple regression analysis, is a strong indication of the relationship between TST and axonopathy (Table 4).

The present study confirms the utility of MEP to document the MS-related functional disability. Our results highlight the additional value provided by TST for the functional quantification of the central motor conduction failure. Even focused on the assessment of the upper limb corticospinal tract, TST appears to reflect a global and degenerative process since it is correlated not only to the grasping strength but also to the walking capacity. In the future, longitudinal studies should be performed to confirm those correlations and investigate the role of TST, alone or in association with demographic, clinical, radiological and conventional electrophysiological data, in order to improve the existing multiparametric prognostic models [42, 43]. TST could be a useful surrogate marker of motor central axonopathy in the era of new very active drugs. The ability to take, not only brain, but also spinal cord lesions into account could improve our decision-making toolbox to evaluate the ability of new drugs to protect axons from inflammatory insults.

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COMPLIANCE WITH ETHICAL STANDARDS

Conflict of interest The authors of this study declare that they have no conflict of interest.

Ethical approval All procedures performed in this study were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Informed consent All patients gave their written informed consent.

References

1. Chen R, Cros D, Curra A, Di Lazzaro V, Lefaucheur JP, Magistris MR, Mills K, Rosler KM, Triggs WJ, Ugawa Y, Ziemann U (2008) The clinical diagnostic utility of transcranial magnetic stimulation: report of an IFCN committee. *Clin Neurophysiol* 119(3):504–532. <https://doi.org/10.1016/j.clinph.2007.10.014>
2. Fernandez V, Valls-Sole J, Relova JL, Raguer N, Miralles F, Dinca L, Taramundi S, Costa-Frossard L, Ferrandiz M, Ramio-Torrenta L, Villoslada P, Saiz A, Calles C, Antigüedad A, Alvarez-Cermeno JC, Prieto JM, Izquierdo G, Montalban X, Fernandez O (2013) Recommendations for the clinical use of motor evoked potentials in multiple sclerosis. *Neurologia* 28(7):408–416. <https://doi.org/10.1016/j.nrl.2012.07.007>
3. Hutchinson M (2013) Evoked potentials are of little use in the diagnosis or monitoring of MS: commentary. *Mult Scler* 19(14):1824–1825. <https://doi.org/10.1177/1352458513506506>
4. Zivadinov R, Leist TP (2005) Clinical-magnetic resonance imaging correlations in multiple sclerosis. *J Neuroimaging* 15(4 Suppl):10S–21S. <https://doi.org/10.1177/1051228405283291>
5. Ingram DA, Thompson AJ, Swash M (1988) Central motor conduction in multiple sclerosis: evaluation of abnormalities revealed by transcutaneous magnetic stimulation of the brain. *J Neurol Neurosurg Psychiatry* 51(4):487–494

6. Kandler RH, Jarratt JA, Davies-Jones GA, Gumpert EJ, Venables GS, Sagar HJ, Zeman A (1991) The role of magnetic stimulation as a quantifier of motor disability in patients with multiple sclerosis. *J Neurol Sci* 106(1):31–34
7. Facchetti D, Mai R, Micheli A, Marciano N, Capra R, Gasparotti R, Poloni M (1997) Motor evoked potentials and disability in secondary progressive multiple sclerosis. *Can J Neurol Sci* 24(4):332–337
8. Gagliardo A, Galli F, Grippo A, Amantini A, Martinelli C, Amato MP, Borsini W (2007) Motor evoked potentials in multiple sclerosis patients without walking limitation: amplitude vs. conduction time abnormalities. *J Neurol* 254(2):220–227. <https://doi.org/10.1007/s00415-006-0334-5>
9. Kale N, Agaoglu J, Onder G, Tanik O (2009) Correlation between disability and transcranial magnetic stimulation abnormalities in patients with multiple sclerosis. *J Clin Neurosci* 16(11):1439–1442. <https://doi.org/10.1016/j.jocn.2009.03.009>
10. Hess CW, Mills KR, Murray NM, Schrieffer TN (1987) Magnetic brain stimulation: central motor conduction studies in multiple sclerosis. *Ann Neurol* 22(6):744–752. <https://doi.org/10.1002/ana.410220611>
11. Kalkers NF, Strijers RL, Jasperse MM, Neacsu V, Geurts JJ, Barkhof F, Polman CH, Stam CJ (2007) Motor evoked potential: a reliable and objective measure to document the functional consequences of multiple sclerosis? Relation to disability and MRI. *Clin Neurophysiol* 118(6):1332–1340. <https://doi.org/10.1016/j.clinph.2007.02.018>
12. Humm AM, Magistris MR, Truffert A, Hess CW, Rosler KM (2003) Central motor conduction differs between acute relapsing-remitting and chronic progressive multiple sclerosis. *Clin Neurophysiol* 114(11):2196–2203
13. Humm AM, Z'Graggen WJ, Buhler R, Magistris MR, Rosler KM (2006) Quantification of central motor conduction deficits in multiple sclerosis patients before and after treatment of acute exacerbation by methylprednisolone. *J Neurol Neurosurg Psychiatry* 77(3):345–350. <https://doi.org/10.1136/jnnp.2005.065284>
14. Comi G, Leocani L, Medaglini S, Locatelli T, Martinelli V, Santuccio G, Rossi P (1999) Evoked potentials in diagnosis and monitoring of multiple sclerosis. *Electroencephalogr Clin Neurophysiol Suppl* 49:13–18
15. Filippi M, Charil A, Rovaris M, Absinta M, Rocca MA (2014) Insights from magnetic resonance imaging. *Handb Clin Neurol* 122:115–149. <https://doi.org/10.1016/B978-0-444-52001-2.00006-6>
16. Rosler KM, Petrow E, Mathis J, Aranyi Z, Hess CW, Magistris MR (2002) Effect of discharge desynchronization on the size of motor evoked potentials: an analysis. *Clin Neurophysiol* 113(11):1680–1687
17. Magistris MR, Rosler KM, Truffert A, Myers JP (1998) Transcranial stimulation excites virtually all motor neurons supplying the target muscle. A demonstration and a method improving the study of motor evoked potentials. *Brain* 121(Pt 3):437–450
18. Banach M, Rakowicz M (2010) Electrophysiological diagnosis of amyotrophic lateral sclerosis. *Przegl Lek* 67(9):736–740
19. Xu Y, Zhang S, Fan D (2015) Upper motor neuron involvement in Kennedy disease evaluated by triple stimulation technique. *Zhonghua Yi Xue Za Zhi* 95(19):1522–1525
20. Sakuma K, Adachi Y, Fukuda H, Kai T, Nakashima K (2005) Triple stimulation technique in patients with spinocerebellar ataxia type 6. *Clin Neurophysiol* 116(11):2586–2591. <https://doi.org/10.1016/j.clinph.2005.04.015>
21. Eusebio A, Azulay JP, Witjas T, Rico A, Attarian S (2007) Assessment of cortico-spinal tract impairment in multiple system atrophy using transcranial magnetic stimulation. *Clin Neurophysiol* 118(4):815–823. <https://doi.org/10.1016/j.clinph.2007.01.004>
22. Attarian S, Franques J, Elisabeth J, Trebuchon A, Duclos Y, Wybrecht D, Verschueren A, Salort-Campana E, Pouget J (2015) Triple-stimulation technique improves the diagnosis of chronic inflammatory demyelinating polyradiculoneuropathy. *Muscle Nerve* 51(4):541–548. <https://doi.org/10.1002/mus.24352>

23. Taieb G, Grapperon AM, Duclos Y, Franques J, Labauge P, Renard D, Yuki N, Attarian S (2015) Proximal conduction block in the pharyngeal-cervical-brachial variant of Guillain–Barre syndrome. *Muscle Nerve* 52(6):1102–1106. <https://doi.org/10.1002/mus.24729>
24. Humm AM, Z'Graggen WJ, von Hornstein NE, Magistris MR, Rosler KM (2004) Assessment of central motor conduction to intrinsic hand muscles using the triple stimulation technique: normal values and repeatability. *Clin Neurophysiol* 115(11):2558–2566. <https://doi.org/10.1016/j.clinph.2004.06.009>
25. Hofstadt-van Oy U, Keune PM, Muenssinger J, Hagenburger D, Oschmann P (2015) Normative data and long-term test–retest reliability of the triple stimulation technique (TST) in multiple sclerosis. *Clin Neurophysiol* 126(2):356–364. <https://doi.org/10.1016/j.clinph.2014.05.032>
26. Rosler KM, Scheidegger O, Magistris MR (2009) Corticospinal output and loss of force during motor fatigue. *Exp Brain Res* 197(2):111–123. <https://doi.org/10.1007/s00221-009-1897-z>
27. Scheidegger O, Kamm CP, Humpert SJ, Rosler KM (2012) Corticospinal output during muscular fatigue differs in multiple sclerosis patients compared to healthy controls. *Mult Scler* 18(10):1500–1506. <https://doi.org/10.1177/1352458512438722>
28. Humm AM, Beer S, Kool J, Magistris MR, Kesselring J, Rosler KM (2004) Quantification of Uhthoff's phenomenon in multiple sclerosis: a magnetic stimulation study. *Clin Neurophysiol* 115(11):2493–2501. <https://doi.org/10.1016/j.clinph.2004.06.010>
29. Firmin L, Muller S, Rosler KM (2012) The latency distribution of motor evoked potentials in patients with multiple sclerosis. *Clin Neurophysiol* 123(12):2414–2421. <https://doi.org/10.1016/j.clinph.2012.05.008>
30. Magistris MR, Rosler KM, Truffert A, Landis T, Hess CW (1999) A clinical study of motor evoked potentials using a triple stimulation technique. *Brain* 122(Pt 2):265–279
31. Polman CH, Reingold SC, Banwell B, Clanet M, Cohen JA, Filippi M, Fujihara K, Havrdova E, Hutchinson M, Kappos L, Lublin FD, Montalban X, O'Connor P, Sandberg-Wollheim M, Thompson AJ, Waubant E, Weinshenker B, Wolinsky JS (2011) Diagnostic criteria for multiple sclerosis: 2010 revisions to the McDonald criteria. *Ann Neurol* 69(2):292–302. <https://doi.org/10.1002/ana.22366>
32. Fischer JS, Jack AJ, Kniker JE, Rudick RA, Cutter G (2001) Administration and scoring manual for multiple sclerosis functional composite (MSFC) (revised). National Multiple Sclerosis Society, New York
33. Roberts HC, Denison HJ, Martin HJ, Patel HP, Syddall H, Cooper C, Sayer AA (2011) A review of the measurement of grip strength in clinical and epidemiological studies: towards a standardised approach. *Age Ageing* 40(4):423–429. <https://doi.org/10.1093/agein/gaf051>
34. Buhler R, Magistris MR, Truffert A, Hess CW, Rosler KM (2001) The triple stimulation technique to study central motor conduction to the lower limbs. *Clin Neurophysiol* 112(5):938–949
35. Magistris RM (2016) The triple stimulation technique: an advanced neurophysiological method to assess motor function in multiple sclerosis. *J Mult Scler* 3:3. <https://doi.org/10.4172/2376-0389.1000188>
36. Fuhr P, Borggrefe-Chappuis A, Schindler C, Kappos L (2001) Visual and motor evoked potentials in the course of multiple sclerosis. *Brain* 124(Pt 11):2162–2168
37. Schmierer K, Irlbacher K, Grosse P, Roricht S, Meyer BU (2002) Correlates of disability in multiple sclerosis detected by transcranial magnetic stimulation. *Neurology* 59(8):1218–1224
38. Leocani L, Rovaris M, Boneschi FM, Medaglini S, Rossi P, Martinelli V, Amadio S, Comi G (2006) Multimodal evoked potentials to assess the evolution of multiple sclerosis: a longitudinal study. *J Neurol Neurosurg Psychiatry* 77(9):1030–1035. <https://doi.org/10.1136/jnnp.2005.086280>
39. van der Kamp W, Maertens de Noordhout A, Thompson PD, Rothwell JC, Day BL, Marsden CD (1991) Correlation of phasic muscle strength and corticomotoneuron conduction time in multiple sclerosis. *Ann Neurol* 29(1):6–12. <https://doi.org/10.1002/ana.410290104>
40. Rocca MA, Horsfield MA, Sala S, Copetti M, Valsasina P, Mesaros S, Martinelli V, Caputo D, Stosic-Opincal T, Drulovic J, Comi G, Filippi M (2011) A multicenter assessment of cervical cord atrophy among MS clinical phenotypes. *Neurology* 76(24):2096–2102. <https://doi.org/10.1212/WNL.0b013e31821f46b8>
41. Boniface SJ, Mills KR, Schubert M (1991) Responses of single spinal motoneurons to magnetic brain stimulation in healthy subjects and patients with multiple sclerosis. *Brain* 114(Pt 1B):643–662

42. Giffroy X, Maes N, Albert A, Maquet P, Crielaard JM, Dive D (2016) Multimodal evoked potentials for functional quantification and prognosis in multiple sclerosis. BMC Neurol 16:83. <https://doi.org/10.1186/s12883-016-0608-1>
43. Giffroy X, Maes N, Albert A, Maquet P, Crielaard JM, Dive D, Humm AM (2016) Do evoked potentials contribute to the functional follow-up and clinical prognosis of multiple sclerosis? Acta Neurol Belg. <https://doi.org/10.1007/s13760-016-0650-1>