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Prospective, blind study of the triple stimulation technique in the diagnosis of ALS

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Abstract

Objective: To evaluate the diagnostic yield of magnetic cortical stimulation with the triple stimulation technique (TST) to identify upper motor neuron (UMN) involvement in patients suspected of having ALS. *Methods:* Fifty-nine patients were recruited to undergo TST in addition to the standard work-up for suspected motor neuron disease. TST combines transcranial magnetic stimulation of the motor cortex with collision studies, which results in a higher sensitivity in detecting UMN involvement. Primary outcome was the number of abnormal TST results in patients with possible ALS. The positivity rate was converted to the number needed to test with TST (NN-TST) for one extra diagnosis of ALS. *Results:* Fifty patients underwent TST. In the total group ($n = 59$), 18 patients had a motor neuron disorder but did not fulfil criteria for 'probable' or 'definite' ALS. In four of these patients TST was abnormal (NN-TST, 4.5). One TST was erroneously interpreted as abnormal. TST findings were normal in inclusion body myositis and peripheral nerve disorders. *Conclusion:* This prospective and blind study confirms open studies of TST in the evaluation of ALS. We suggest that TST can be used to arrive at a diagnosis of 'probable' or 'definite' ALS in patients lacking UMN signs in the upper extremities.

Key words: *Upper motor neuron signs, transcranial magnetic stimulation, diagnostic study, STARD*

Introduction

Amyotrophic lateral sclerosis is the neurodegenerative disorder defined by progressive loss of motor neurons in the motor cortex, brainstem, and spinal cord (1). The diagnosis requires signs of upper and lower motor neuron (UMN, LMN) loss, with diagnostic certainty depending on the extent of signs (2,3). The spread of abnormality to the different regions of the body, defined in the El Escorial and Airlie House criteria of diagnostic certainty, correlates negatively with the number of diagnostic errors (4–6). However, the classification is not related to severity of the disease, as clearly indicated by the fact that survival is comparable in 'possible' and 'definite' ALS (7). In the early diagnosis of ALS a change from 'possible' to 'probable' ALS is relevant in the first place because a clear diagnosis is important for the patient and may improve communication of the diagnosis (8,9). Secondary to a clear diagnosis, neuroprotective therapy with riluzole can be started early and treatment trials may be more successful before neurodegeneration is widespread (7,10,11).

EMG can supplement clinical evaluation in the detection of LMN deficits when diagnosing 'probable laboratory supported' ALS, and further revision of criteria has been proposed recently (12). The application of the Awaji algorithm, which puts more emphasis on the EMG in general and fasciculation potentials in particular, improves diagnosis of LMN abnormalities (13). However, the detection of UMN deficits is currently based on clinical examination alone. Transcranial magnetic stimulation may be used to measure the function of UMN, but the most frequently used measures are not very sensitive for UMN deficits that are not clinically apparent. Therefore, such techniques are not currently mentioned in diagnostic criteria (12,14). For diagnostic purposes, the limiting factor is the large variation of

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motor evoked potentials (MEPs) within and between subjects. This variation is due not to submaximal stimulation, but to the variable timing of action potentials resulting in dispersion and phase cancellation (15). Magistris et al. combined transcranial magnetic stimulation with a collision technique that solves several of these problems with amplitude measurements. With their triple stimulation technique (TST), response amplitude directly reflects the motor units and muscle fibres that can be activated from the cortex. Reference values for TST have a narrow range that results in higher sensitivity in the detection of UMN involvement in a number of central motor disorders, including ALS (16,17). TST results correlate to some extent with the severity of the UMN deficit and are more often abnormal in patients with 'definite' or 'probable' ALS than in patients with 'possible' ALS (17,18). In patients already diagnosed with ALS based on signs in another region, TST may identify a previously undetected abnormality of the UMN (16,18–20). On the basis of these results, it has been suggested that TST can increase diagnostic certainty in ALS, but to date the method has not been evaluated in a prospective study.

We expected that TST would detect UMN deficits in some patients in whom muscle tone and reflexes in the upper extremity were normal on clinical evaluation. Thus, TST findings, in combination with other signs and findings, may increase diagnostic certainty, for example by grading up from 'possible' ALS to 'probable' ALS. In analogy to a therapeutic trial, we calculated the 'number needed to test' (21,22) to increase the diagnostic certainty in one patient. The Standards for Reporting of Diagnostic accuracy (STARD) checklist was applied (23).

Methods

Patients

All patients referred to our tertiary neuromuscular centre for evaluation of suspected motor neuron disorders were eligible for inclusion in the study. Motor neuron disease had to be considered in the differential diagnosis, but not necessarily as the most likely diagnosis. Exclusion criteria were seizures, and metallic or electronic implants. All patients gave informed consent. The study protocol was approved by the local committee on research involving human subjects.

Triple stimulation technique: set-up

For a complete theoretical and physiological background of the method we refer to the initial report by Magistris et al. (15). Briefly, the first magnetic stimulus induces action potentials in the cortex that travel along the corticospinal tract and the peripheral nerve. The other two stimuli, applied at the wrist and at Erb's point, are set up to quantify the result of the first stimulus and to remove any dispersion. Comparison of TST after cortical stimulation (TST test) with that after Erb's stimulation (TST control) gives a direct estimate of UMN function. In the following, we give the full technical details of our implementation.

Self-adhesive electrodes (22 × 22 mm Ag/AgCl, Kendall H69P, Tyco Healthcare, Mansfield, MA, USA) were placed at the motor point of the abductor digiti minimi muscle and at the proximal phalanx of the little finger. The EMG signal was amplified (250x, Hydiak 693 DC/AC, EKIDA, Helmstadt, Germany), filtered (10–3000 Hz), and digitized (10 kHz, 0.6 μV/bit, CED Power 1401, Cambridge Electronic Design, Cambridge, UK). Data acquisition, on-line visualization, and stimulus timing were controlled with Spike2 software. Patients received visual feedback of the baseline-corrected, rectified, and smoothed (100 ms moving average) EMG at 200 ms/cm. Post-stimulus data were displayed at 2.5 ms/cm (0–50 ms). Default display gain was 3 mV/cm for adjustment of amplitude markers and 200 μV/cm for the placement of latency markers.

For peripheral nerve stimulation, a pair of self-adhesive electrodes (3M Red Dot 2271, 3M Nederland, Zouterwoude, NL) was placed with the cathode just proximal to the wrist over the ulnar nerve and the anode medial and slightly proximal to it. The brachial plexus was stimulated with large electrodes (5 × 4.5 cm, Thymapad ECT stimulus electrodes, Somatics LLC, Lake Bluff, IL, USA) with the cathode at Erb's point and the anode at the scapula (24). Electrodes were connected to constant current stimulators (DS7A and DS7AH, Digitimer, Welwyn Garden City, UK). By using a high-voltage stimulator, supra-maximal stimulation was achieved with a pulse duration of 0.1 ms. The motor cortex was stimulated with a magnetic stimulator (Magstim 200, Magstim, Witland, UK) connected to a 90-mm circular coil centered above the vertex. Stimulation intensity was 80% or 100% of maximum stimulator output.

Triple stimulation technique: measurement protocol

During the measurements, the subject was lying on a bed in supine position. To avoid movement artifacts, the fingers were fixed with tape. The measurements started with three recordings of maximum voluntary abduction of the little finger, with the patients receiving EMG feedback. The highest stable amplitude was selected and 20% of the maximum EMG was calculated as the target level for tonic voluntary contraction. Supra-maximum stimulation intensity was adjusted for ulnar nerve stimulation and proximal stimulation during rest. The onset latency of the compound muscle action potential (CMAP) was

measured. Its amplitude was measured from baseline to negative peak and was required to exceed 2.0 mV.

During tonic contraction at 20% maximum EMG, three to five magnetic stimuli were given and the MEP was recorded. The latency of the MEP was measured. The absence of a MEP in response to cortical stimulation was considered as abnormal, equivalent to an abnormal TST result and indicative of a UMN lesion.

To achieve appropriate collision of action potentials, the timing of TST was automatically adjusted according to the conduction time of the patient. Delay I was calculated as the difference between MEP latency and distal motor latency. Delay II was the difference between CMAP latency in response to Erb's points stimulation and distal motor latency. As defined by Magistris et al., distal motor latency was rounded up to the next millisecond, while MEP latency and Erb latency were rounded down to the nearest millisecond. The first and the last TST trials were given as TST control stimulations. In these trials, Erb's stimulation was followed after delay II by wrist stimulation and then again by Erb's stimulation after delay II. TST test stimulation consisted of motor cortex stimulation, wrist stimulation after delay I, and Erb's stimulation after delay II. Three TST test trials were performed, with the magnetic stimulator set to 80%, 100%, and 100% of maximum stimulator output. No threshold measurements were performed. All five trials were superimposed to check for signal quality, movement artifacts, and consistent supramaximal stimulation (Figure 1).

As voluntary pre-activation may be required for maximum activation of the hand muscles, all TST trials were performed with 20% voluntary contraction. A sandbag was used to avoid changes in hand position between trials. The number of TST trials was predefined to exclude that the investigation was repeated until the investigator was 'satisfied'.

The amplitude of the TST response was measured from baseline to the negative peak. TST amplitude ratio was defined as the ratio between the maximum of three TST test amplitudes divided by the maximum of two TST control amplitudes. A TST amplitude ratio of less than 90% was considered abnormal (18). In the literature, 93% was established as the lower (2.5 SD) limit of normal and was used as the diagnostic cut-off value. Because of the predefined and low number of trials, our

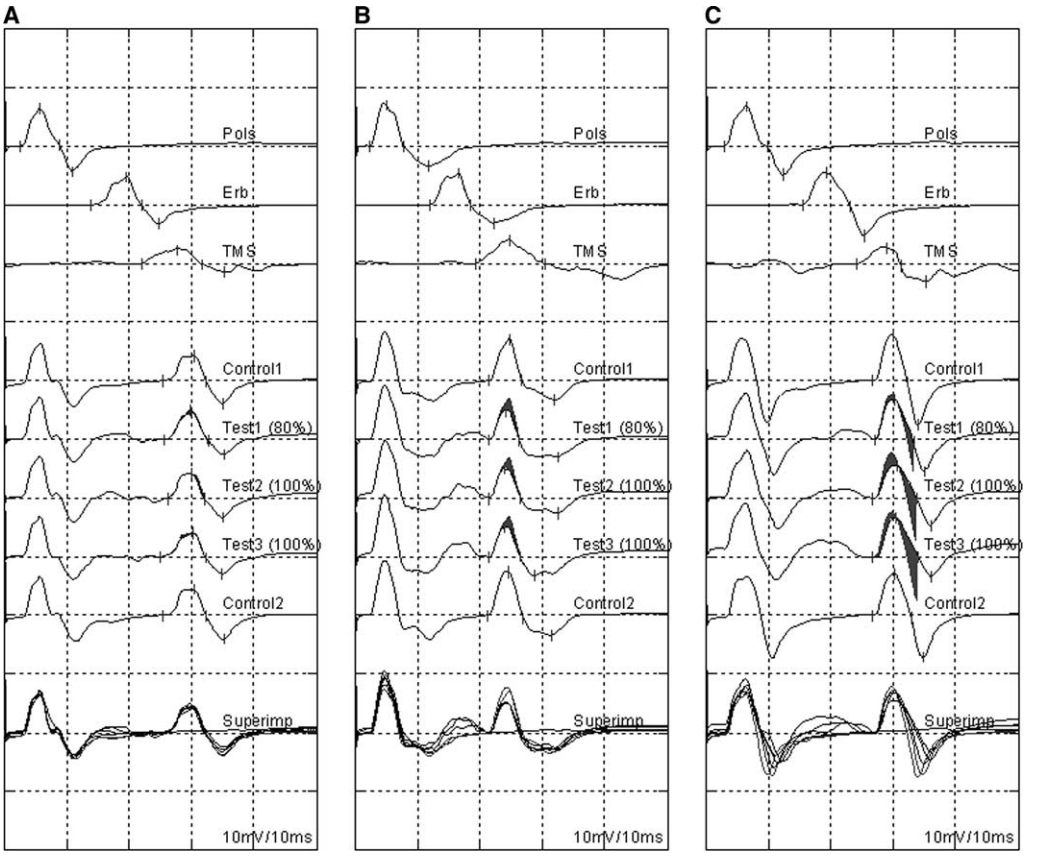

Figure 1. Examples of TST curves as they appear in the report. The first three traces give the response to separate stimulation of ulnar nerve, plexus brachialis and motor cortex. The following five traces give the TST control and TST test curves. The difference between first TST control and the TST test curves is filled. In the lowest traces TST test curves (black) and TST control curves (grey) are superimposed. (A) shows a normal TST amplitude ratio. In (B) TST is abnormal with an amplitude ratio of 69%. In (C) the hand position had changed between measurements, as can be seen from the shorter duration of the CMAP and the TST response. This artifact was missed initially and only recognized later (see text). TST amplitude ratio was 87%.

cut-off was set lower than recommended initially, but comparable to that for the first dorsal interosseus muscle (15,25). Inter-session and inter-reader agreement of the method are reported to be within about 6% (15,25). An example of a normal and an abnormal TST result is given in Figure 1.

Study design

Between October 2006 and October 2007, consecutive patients were included prospectively provided that TST could be performed on the same day as the neurological consultation and EMG. Demographic data, symptom duration, and the presence of UMN or LMN signs per region were recorded by a neurologist (HJS). Disease duration was determined from when first symptoms were apparent, as indicated in the patient's history. Nerve conduction studies and needle EMG were performed by a neurologist specialized in neuromuscular disease and motor neuron disorders (HJS) or by a neurology resident supervised by a clinical neurophysiologist (MJZ). Arlie House criteria for ALS (3) were applied by the neurologist (HJS), who did not know the TST results. All patients with LMN signs only were classified as having progressive muscular atrophy (PMA), which overlaps with 'suspected ALS' in the El Escorial classification (2,26,27).

TST was performed and read by a resident in neurology and clinical neurophysiology (BUK), together with a technician. The results of neurological examination and EMG were not available during TST. Independent of clinical presentation, both hands were tested and a TST amplitude ratio of less than 90% was considered abnormal (18). If one or both sides were abnormal, the TST findings were reported as an indication of UMN deficit in muscles of the cervical region. At the time of diagnosis, the neurologist and clinical neurophysiologist were blind to the results of TST.

If ALS was the initial diagnosis or no alternative diagnosis was established, patients were followed up for at least six months. The neurologist re-assessed the patient and ordered additional tests as required by the clinical situation. Progression, as expected from the previous history, was required to confirm the diagnosis. According to national guidelines, patients were referred to the closest possible multidisciplinary ALS team. In patients that were not re-assessed at six months at our department, progression on follow-up by another ALS team was considered equivalent. The neurologist and the ALS teams had no access to the TST results at follow-up.

Outcome measures and statistics

The primary outcome measure was the positivity rate, i.e. the number of patients that changed in diagnostic category from 'PMA/suspected' or 'possible' or 'probable laboratory supported' ALS to 'probable' or 'definite' on the basis of an abnormal TST ratio. Progression on follow-up was the reference standard. We compared the number of patients who received a diagnosis of 'probable' or 'definite' ALS on clinical grounds with the number of patients who would have such a diagnosis after TST. In clinical trials the effect of treatment can be measured by the number needed to treat, i.e. the number of patients that must receive treatment to achieve one extra favourable outcome (21). Using the same formula, the number needed to test with TST (NN-TST) indicates how many patients need to undergo TST to diagnose one extra case of ALS with the required certainty (22,28). From the standard error of the absolute risk difference the 95% confidence interval of the NN-TST was estimated (29,30). The NN-TST was calculated for the group of patients in a low El-Escorial category and for the whole patient group. NN-TST as a primary outcome measure was calculated for the intention-to-test population, i.e. assuming a normal TST ratio in all patients who have not completed the TST protocol. As a secondary outcome, NN-TST was calculated in the patient group with complete and interpretable TST measurements ('per protocol' analysis). From the false-positive rate the number needed to harm (by incorrectly diagnosing ALS) with TST was calculated. Analysis was on the patient level (31). A priori, no formal calculation of study size was performed. However, an analysis of outcomes was scheduled after 50 patients.

Results

Patient group

As summarized in Figure 2, 59 patients consented to participate. Their demographic characteristics and disease duration are given in Table I. After work-up for ALS, 30 patients were diagnosed to have a variant of motor neuron disease; 10, other central motor disorders; 17, peripheral nerve disorder; and two, no neurological disease.

Fifty patients underwent TST. One patient was excluded from TST because of a cardiac pacemaker, one patient had a history of ruptured aneurysm that was treated with a potentially magnetic clip, six patients stopped early in the recording either due to the unpleasant sensation of magnetic stimulation or due to pain from electrical stimulation at Erb's point. In one patient severe dyspnoea precluded TST. Only one patient could not tolerate TST at the other side after unilateral measurement. His TST ratio was assumed to be normal for further analysis.

In five patients a TST amplitude ratio could not be obtained because of peripheral abnormalities. In three of these patients conduction block was

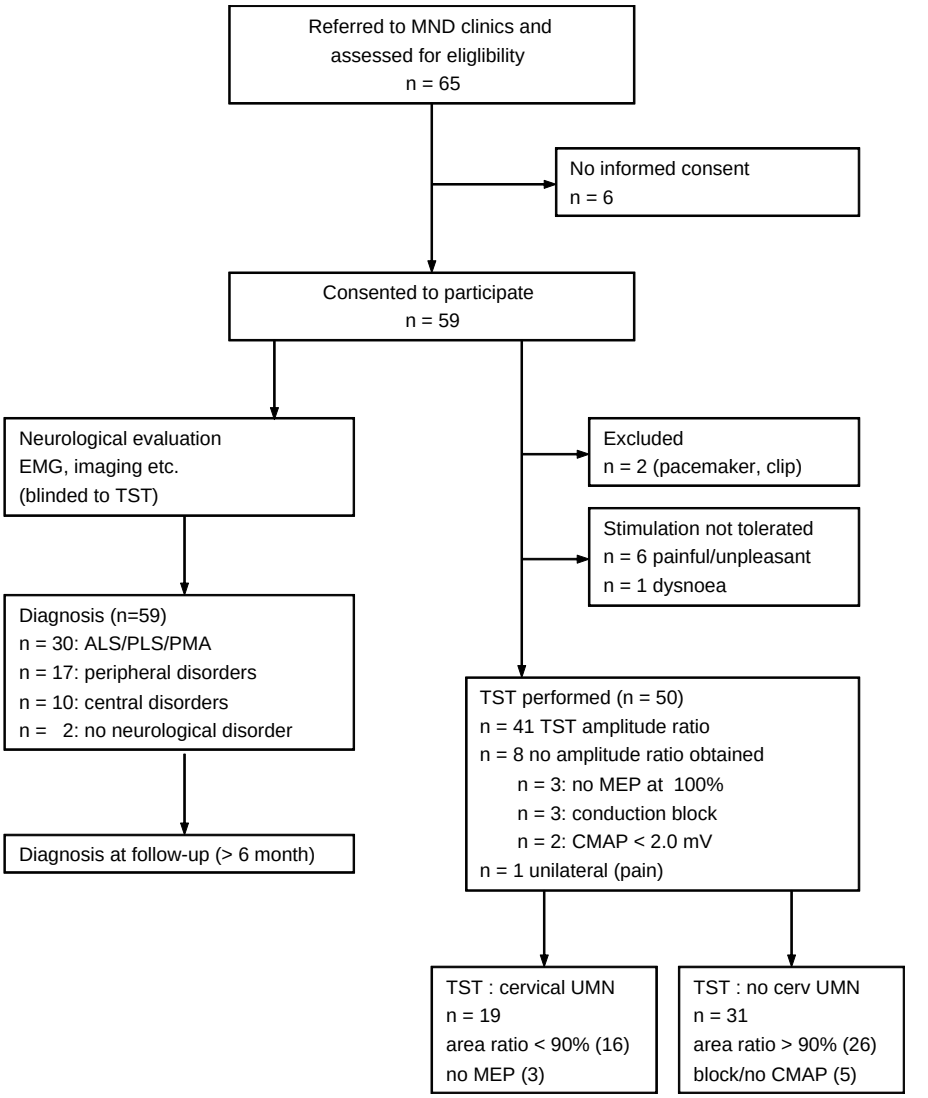

Figure 2. Flow diagram summarizing prospective inclusion of patients in the study. The left side gives the diagnostic process. On the right side the results of the TST study are summarized. Note that the diagnosis and follow-up was blinded for the TST results.

detected and blinding was broken. In another three patients no MEP and therefore also no TST test response was obtained. These patients were considered to have a lesion of the UMNs innervating the cervical myotomes.

During follow-up, one patient showed no progression and cramps improved; the diagnosis was changed from PMA to benign fasciculations and cramps. Another patient progressed from PMA to probable ALS. All patients diagnosed with 'possible', 'probable laboratory supported', 'probable' or 'definite' ALS showed some degree of symptom progression.

Primary outcome measures

In the group with motor neuron disease ($n = 30$), 18 patients did not fulfil the criteria for 'probable' or 'definite' ALS. Of these, 16 patients would have shifted to a higher diagnostic category if a UMN lesion in the cervical region were found (Table II). In four of them TST amplitude ratio was abnormal, resulting in a positivity rate of 4/18 corresponding to a NN-TST of 4.5 with a confidence interval not including infinity (CI 2.4–33). There were six patients with UMN signs in only one region of the body (Table II, 'probable laboratory supported'). Of these six patients, five had the UMN signs in the legs or in the bulbar region only. If TST found evidence of abnormality in the UMNs supporting the cervical region, two body regions could be classified as abnormal, resulting in 'probable' ALS. In the sixth patient, UMN signs were present in the upper extremity, such that TST would not add any new information. In four patients TST was performed 'per protocol', three times with a TST amplitude ratio of less than 90% (Table II, first row). For the whole patient population, four of 59 patients changed category, resulting in a NN-TST of 15. The confidence interval was wide and included infinity (CI 4.8 to ∞ and $-\infty$ to -13).

Table I. Patient population.

	Age (y) (median, range)	Duration (m) (median, range)	Region of onset (b/c/l)
All patients ($n=59$)	58 (23–81)	16 (2–300)	4/27/27
Definite ALS (2)	(51, 60)	(8, 35)	(2/–/–)
Probable ALS (8)	66 (40–71)	9 (5–44)	(2/3/3)
Probable lab. supp. ALS (6)	56 (46–81)	18 (6–37)	(–/5/1)
Possible ALS (4)	47 (32–58)	7 (6–114)	(–/2/2)
PMA/suspected ALS (8)	61 (45–77)	25 (5–121)	(–/5/3)
PLS (2)	(46, 63)	(11, 61)	(–/1/l)
Vascular (4)*	71 (64–74)	100 (26–156)	(–/–/4)
HSP (4)	38 (23–41)	66 (12–299)	(–/–/4)
MSA (2)	(70, 82)	(30,132)	(–/–/1)
Radiculopathy **(3)	65 (56–73)	24 (9–97)	(–/2/1)
IBM (3)	63 (55–66)	48 (12–75)	(–/1/2)
Neuropathy *** (3)	67 (59–75)	9 (6–11)	(–/1/2)
MMN (2)	(38,49)	(15,53)	(–/2/–)
Ulnar neuropathy (2)	(59, 70)	(4, 50)	(–/2/–)
Benign fasciculations ****(3)	55 (49–58)	16 (2,19)	(–/1/2)
Radiation plexopathy (1)	70	7	(–/1/–)
No neurological dis. (2)	(41,48)	(16,60)	(–/1/1)

* Stroke or extensive white matter lesions on cerebral imaging.
** In one patient myelopathy in combination with radiculopathy.
*** Diabetic, hereditary sensory and motor neuropathy, sequelae of Guillain-Barré syndrome.
**** In one patient benign fasciculations and a history of cerebral hypoxia due to cardiac arrhythmia.

In one patient, diagnosed with spondylotic lumbosacral polyradiculopathy, a TST amplitude ratio of 87% was obtained on stimulation of the left side (Figure 1C). Assuming that a false diagnosis of ALS may have been given to this patient, the number needed to harm would be 13 (lower limit of 95% CI, 4.8). In this calculation a 'worst case' is considered. One has to assume that the patient belonged to the per protocol group of 12 patients and that one extra patient received a (false-positive) diagnosis of ALS. We later discovered that the patient's hand had changed position between tests, changing the shape of the action potential.

Secondary outcome measures

Of the 10 patients with 'probable laboratory supported', 'possible' or 'suspected' ALS and complete TST data, the positivity rate was 4/10. This means that four patients changed diagnostic category on the basis of the TST findings, corresponding to a per protocol NN-TST of 2.5 (CI 1.4–10) for low El Escorial categories (Figure 3). For the group of 50 patients who underwent TST, the per protocol NN-TST was 13 (CI, 4 to ∞ and $-\infty$ to -12).

Overall, of the 50 TST performed, results were abnormal in 19 patients. Of these, 14 were in patients with known or suspected central motor disorders (Table III, Figure 3). This group included patients with UMN signs sufficient to diagnose 'probable' or 'definite' ALS, but also other diseases that were suspected from history and confirmed by other parts of the neurological examination or by neuroimaging. One patient had severe UMN signs in all regions of the body with only minor LMN involvement, resulting in a diagnosis of 'possible' ALS (Table III). TST added no truly new diagnostic information in these patients. Except for one false-positive result (see above), the TST amplitude ratio was normal in inclusion body myositis and peripheral nerve disorders. In two patients, one with 'probable ALS' and one with hereditary spastic paraparesis (HSP), the TST amplitude ratio was between the two recommended cut-off points of 90% and 93% (15,18).

Discussion

To our knowledge, this is the first prospective, blind study of the use of TST for the detection of

Table II. TST results in patients in a low category of the El Escorial criteria.

Diagnosis	Total (intention-to-test)	Could change category	TST (per protocol)	TST ampl. ratio (mean)	TST abnormal
Probable lab. supp. ALS	6	5	4	84%	3
Possible ALS	4	3	3	95%	1
PMA/suspected ALS	8	8	3	105%	0
	18	16	10	94%	4

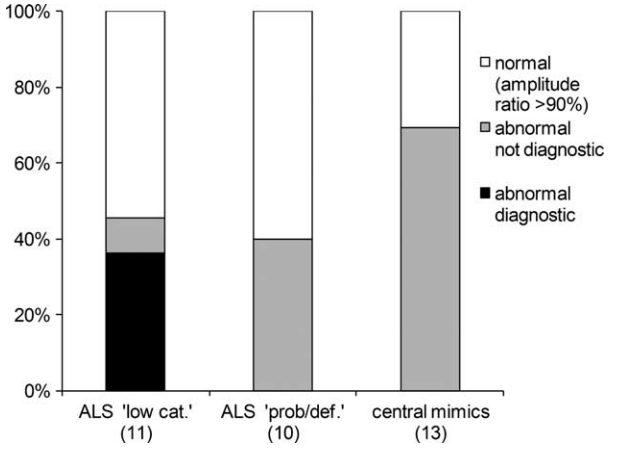

Figure 3. Percentage of abnormal TST amplitude ratios. The number of abnormal tests is similar in the group with 'probable/definite' ALS and in the group with 'probable laboratory supported' ALS or lower. In the lower diagnostic category, most abnormal TST results contributed to diagnostic certainty (black bar).

subclinical UMN abnormalities in ALS. TST findings were abnormal in about one in five patients who ultimately developed ALS but who did not meet the diagnostic criteria for 'definite' or 'probable' ALS. The positivity rate was 4/18 and the NN-TST was 4.5 (CI 2.4–33). The lack of infinity in the confidence interval indicates that TST adds a statistically significant amount of diagnostic information. This was also the case for the intention-to-test analysis, indicating that even with a small number of drop-outs the statistically significant advantage was retained.

Previous studies of TST in patients with ALS (16–18,20) were not blinded, so the clinical diagnosis may have been influenced by knowledge of TST results. However, the number of abnormal TST results in our study was comparable to that reported in earlier studies, i.e. this study replicates the diagnostic yield of TST in a blinded setting. As in open studies, the TST amplitude ratio was more often abnormal in patients with clear UMN signs, indicating spectrum bias (17,18,32,33). Thus, TST is most useful in patients with minor UMN involvement, in whom the findings of the neurological examination are still normal. In these patients, TST results are abnormal, but are often relatively close to the limit of normal. Although most patients with 'possible' or 'probable laboratory supported' ALS will eventually develop ALS, it is important to reduce uncertainty about the diagnosis, both for patients and their doctors (8,9).

All patients diagnosed with 'PMA/suspected ALS' had a normal TST amplitude ratio. It remains unclear whether these patients did not have UMN deficits or whether the technique is not sensitive enough to detect them. Changing the cut-off value for an abnormal TST amplitude ratio from 90% to 93% would not solve the problem.

Only the cervical region was investigated in this study, as TST was performed in intrinsic muscles of both hands. A TST technique has been described also for the lower extremity, but its sensitivity in the diagnosis of ALS is unknown (34).

In contrast to previous studies of TST in ALS, this study included all patients referred to a motor neuron disorders clinic. Many of them were ultimately diagnosed with central and peripheral mimics of ALS. Except for one false-positive result due to technical problems, no abnormal TST amplitude ratios were found in patients with peripheral nerve or muscle disorders, indicating a high specificity. In some patients with peripheral abnormality, TST does not give useful results. In case of conduction block between the wrist and Erb's point, the study is diagnostic because ALS is ruled out, but central conduction itself becomes inaccessible. With severe loss of motor units resulting in atrophy (CMAP below 2 mV) or with a single unit pattern, TST amplitude cannot be interpreted (15,18). This will not give any diagnostic problems, as all rules for assessing proximal or central abnormalities in the context of peripheral abnormalities are well established for both nerve conduction studies and the neurological examination.

As expected, a substantial number of abnormal results were obtained in patients with different central motor disorders. Both within and outside the context of ALS an abnormal TST can be interpreted almost in the same way as UMN signs.

The probability of ALS is increased by finding a low TST amplitude ratio, provided that other pathology within the corticospinal system is excluded by appropriate neuroimaging. In this sense, TST is generally not specific for ALS. In particular,

Table III. TST results in patients with known central motor disorders.

Diagnosis	Tested with TST	TST amplitude ratio (mean)	Abnormal TST amplitude ratio
Definite ALS	2	51%	1
Probable ALS	8	87%	3
Possible ALS	1	34%	1
PLS	1	0%	1
Vascular *	5	70%	3
HSP	4	79%	3
MSA	2	90%	1
Myelopathy	1	84%	1

* Including one patient with benign fasciculations and a history of cerebral hypoxia.

this remains true for the differential diagnosis between ALS and cervical spondylotic myelopathy. Some additional information can be gained from the central motor conduction time that is more often abnormal in the case of compression (17). However, the contribution of different tests such as conduction time to the hand muscles, to the trapezius muscle or needle EMG of the trapezius muscle has not been studied prospectively (35,36).

In one important aspect a low TST amplitude ratio differs from clinical UMN signs. The TST control test is obtained by stimulating the brachial plexus at Erb's point. Therefore, an abnormality at the root or plexus level cannot be differentiated from abnormality in the central nervous system (15). In some patients with multifocal motor neuropathy, isolated proximal conduction block can be demonstrated with TST or with stimulation at the root level (37–39). In our study, we considered a misdiagnosis unlikely, because even in the absence of conduction block, peripheral nerve conduction studies will reveal some abnormalities that point to the neuropathy.

Given low diagnostic contribution (high NN-TST) for the whole patient population, we think that TST should not be used in every patient referred to the motor neuron clinic. As defined in our primary outcome measures, in patients who are classified with 'possible' or as 'probable laboratory supported', ALS TST can contribute to an early diagnosis by finding subclinical evidence of UMN abnormality. In the Awaji consensus, the 'probable laboratory supported' category is removed, but the need for a test of UMN function may actually increase (12). Using the revised El Escorial criteria together with the new proposal (Awaji algorithm), results in increased sensitivity without loss of specificity (3,13). However, when using the Awaji proposal as a stand-alone set of criteria (i.e. truly removing 'laboratory supported'), this reduces sensitivity for a subgroup of patients, as is best illustrated by an example: Consider a patient with pseudobulbar speech, tongue atrophy and EMG abnormalities including fibrillations in two arm muscles. According to the Airlie House criteria, he or she will be classified as 'probable laboratory supported' ALS. Using the Awaji criteria alone, the classification would be 'possible' ALS, because only one (bulbar) region with UMN abnormality was found. This patient has a three in four chance to have a low TST amplitude ratio in one of the hands (Table III, Figure 3). Equivalence of clinical and neurophysiological abnormality in finding LMN abnormality is one of the principles of the Awaji consensus, but still awaits validation (40). Extending these principles to the UMN, we propose that such a patient is classified as 'probable' ALS. A limitation of our study is the relatively low number of patients. A replication in a larger, preferentially multicentre study would both increase the statistical power and validate our results within the context of the new Awaji criteria.

In conclusion, TST may be a useful tool with a high sensitivity for detecting subclinical UMN abnormalities in suspected ALS. As a result, the level of diagnostic certainty in the evaluation of ALS can be increased.

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References

1. Mitchell JD, Borasio GD. Amyotrophic lateral sclerosis. Lancet. 2007;369:2031–41.
2. Brooks BR. El Escorial World Federation of Neurology criteria for the diagnosis of amyotrophic lateral sclerosis. J Neurol Sci. 1994;124 (Suppl):96–107.
3. Brooks BR, Miller RG, Swash M, Munsat TL. El Escorial revisited: revised criteria for the diagnosis of amyotrophic lateral sclerosis. Amyotroph Lateral Scler Other Motor Neuron Disord. 2000;1:293–9.
4. Traynor BJ, Codd MB, Corr B, Forde C, Frost E, Hardiman OM. Clinical features of amyotrophic lateral sclerosis according to the El Escorial and Airlie House diagnostic criteria: a population based study. Arch Neurol. 2000;57:1171–6.
5. Belsh JM, Schiffman PL. The amyotrophic lateral sclerosis (ALS) patient perspective on misdiagnosis and its repercussions. J Neurol Sci. 1996;139 (Suppl):110–6.
6. Davenport RJ, Swingler RJ, Chancellor AM, Warlow CP. Avoiding false-positive diagnoses of motor neuron disease: lessons from the Scottish Motor Neuron Disease Register. J Neurol Neurosurg Psychiatry. 1996;60:147–51.
7. Zoccolella S, Beghi E, Palagano G, Fraddosio A, Samarelli V, Lamberti P, et al. Predictors of delay in the diagnosis and clinical trial entry of amyotrophic lateral sclerosis patients: a population based study. J Neurol Sci. 2006;250:45–9.
8. Borasio GD, Sloan R, Pongratz DE. Breaking the news in amyotrophic lateral sclerosis. J Neurol Sci. 1998;160 (Suppl):127–33.
9. Borasio GD, Pongratz DE. Gedanken zur Aufklärung bei amyotropher Lateralskleose. Nervenarzt. 1997;68:1004–7.
10. Bensimon G, Lacomblez L, Meininger V, Bouche P, Delwaide C, Couratier P, et al. A controlled trial of riluzole in amyotrophic lateral sclerosis. N Engl J Med. 1994;330: 585–91.
11. Leigh PN, Swash M, Iwasaki Y, Ludolph A, Meininger V, Miller RG, et al. Amyotrophic lateral sclerosis: a consensus viewpoint on designing and implementing a clinical trial. Amyotroph Lateral Scler Other Motor Neuron Disord. 2004;5:84–98.
12. de Carvalho M, Dengler R, Eisen A, England JD, Kaji R, Kimura J, et al. Electrodiagnostic criteria for diagnosis of ALS. Clin Neurophysiol. 2008;119:497–503.
13. de Carvalho M, Swash M. Awaji diagnostic algorithm increases sensitivity of El Escorial criteria for ALS diagnosis. Amyotrophic Lateral Sclerosis. 2009;10:53–7.
14. Chen R, Cros D, Curra A, Di Lazzaro V, Lefaucheur JP, Magistris MR, et al. The clinical diagnostic utility of transcranial magnetic stimulation: report of an IFCN committee. Clin Neurophysiol. 2008;119:504–32.

15. Magistris MR, Rösler KM, Truffert A, Myers JP. Transcranial stimulation excites virtually all motor neurons supplying the target muscle. A demonstration and a method improving the study of motor evoked potentials. Brain. 1998;121: 437–50.
16. Rösler KM, Truffert A, Hess CW, Magistris MR. Quantification of upper motor neuron loss in amyotrophic lateral sclerosis. Clin Neurophysiol. 2000;111:2208–18.
17. Magistris MR, Rösler KM, Truffert A, Landis T, Hess CW. A clinical study of motor evoked potentials using a triple stimulation technique. Brain. 1999;122:265–79.
18. Attarian S, Verschueren A, Pouget J. Magnetic stimulation including the triple stimulation technique in amyotrophic lateral sclerosis. Muscle Nerve. 2007;36:55–61.
19. Rösler KM, Magistris MR. Triple stimulation technique (TST) in amyotrophic lateral sclerosis. Clin Neurophysiol. 2004;115:1715.
20. Komissarow L, Rollnik JD, Bogdanova D, Krampfl K, Khabirov FA, Kossev A, et al. Triple stimulation technique (TST) in amyotrophic lateral sclerosis. Clin Neurophysiol. 2004;115:356–60.
21. Laupacis A, Sackett DL, Roberts RS. An assessment of clinically useful measures of the consequences of treatment. N Engl J Med. 1988;318:1728–33.
22. de Lemos ML. Storm over screening for prostate specific antigen. Number needed to test needs to be known. Br Med J. 2002;324:1392.
23. Bossuyt PM, Reitsma JB, Bruns DE, Gatsonis CA, Glasziou PP, Irwig LM, et al. Towards complete and accurate reporting of studies of diagnostic accuracy: the STARD initiative. Br Med J. 2003;326:41–4.
24. Roth G, Magistris MR. Detection of conduction block by monopolar percutaneous stimulation of the brachial plexus. Electromyogr Clin Neurophysiol. 1987;27:45–53.
25. Humm AM, Z'Graggen WJ, von Hornstein NE, Magistris MR, Rösler KM. Assessment of central motor conduction to intrinsic hand muscles using the triple stimulation technique: normal values and repeatability. Clin Neurophysiol. 2004;115:2558–66.
26. van den Berg-Vos RM, Visser J, Franssen H, de Visser M, de Jong JM, Kalmijn S, et al. Sporadic lower motor neuron disease with adult onset: classification of subtypes. Brain. 2003;126:1036–47.
27. Visser J, van den Berg-Vos RM, Franssen H, van den Berg LH, Wokke JH, de Jong JM, et al. Disease course and prognostic factors of progressive muscular atrophy. Arch Neurol. 2007;64:522–8.
28. Edwards A, Elwyn G, Mulley A. Explaining risks: turning numerical data into meaningful pictures. Br Med J. 2002;324:827–30.
29. Daly LE. Confidence limits made easy: interval estimation using a substitution method. Am J Epidemiol. 1998;147: 783–90.
30. Altman DG. Confidence intervals for the number needed to treat. Br Med J. 1998;317:1309–12.
31. Padua L, Pasqualetti P, Rosenbaum R. One patient, two carpal tunnels: statistical and clinical analysis – by hand or by patient? Clin Neurophysiol. 2005;116:241–3.
32. Ransohoff DF, Feinstein AR. Problems of spectrum and bias in evaluating the efficacy of diagnostic tests. N Engl J Med. 1978;299:926–30.
33. Eusebio A, Azulay JP, Witjas T, Rico A, Attarian S. Assessment of corticospinal tract impairment in multiple system atrophy using transcranial magnetic stimulation. Clin Neurophysiol. 2007;118:815–23.
34. Buhler R, Magistris MR, Truffert A, Hess CW, Rosler KM. The triple stimulation technique to study central motor conduction to the lower limbs. Clin Neurophysiol. 2001;112: 938–49.
35. Truffert A, Rösler KM, Magistris MR. Amyotrophic lateral sclerosis versus cervical spondylotic myelopathy: a study using transcranial magnetic stimulation with recordings from the trapezius and limb muscles. Clin Neurophysiol. 2000;111:1031–8.
36. Sonoo M, Kuwabara S, Shimizu T, Komori T, Hirashima F, Inaba A, et al. Utility of trapezius EMG for diagnosis of amyotrophic lateral sclerosis. Muscle Nerve. 2009;39:63–70.
37. Deroide N, Uzenot D, Verschueren A, Azulay JP, Pouget J, Attarian S. Triple stimulation technique in multifocal neuropathy with conduction block. Muscle Nerve. 2007;35:632–6.
38. Vucic S, Black K, Siao Tick CP, Cros D. Cervical nerve root stimulation. Part II: findings in primary demyelinating neuropathies and motor neuron disease. Clin Neurophysiol. 2006;117:398–404.
39. Arunachalam R, Osei-Lah A, Mills KR. Transcutaneous cervical root stimulation in the diagnosis of multifocal motor neuropathy with conduction block. J Neurol Neurosurg Psychiatry. 2003;74:1329–31.
40. Schelhaas HJ, Kleine BU, Zwarts MJ. Electrodiagnostic criteria for ALS: time to STARD. Clin Neurophysiol. 2008;119:1689–90.