Article:

Capozio, A, Chakrabarty, S and Astill, S (2021) Reliability of the TMS-conditioned monosynaptic reflex in the Flexor Carpi Radialis muscle. Neuroscience Letters, 745. 135622. ISSN: 0304-3940

https://doi.org/10.1016/j.neulet.2020.135622

Reliability of the TMS-conditioned monosynaptic reflex in the Flexor Carpi Radialis muscle

Antonio Capozio[1], Samit Chakrabarty[1], Sarah Astill[1]

[1]School of Biomedical Sciences, University of Leeds, United Kingdom

Corresponding Author: Antonio Capozio

Manton 6.87

School of Biomedical Sciences, University of Leeds

Leeds, LS2 9JT, United Kingdom

E-mail: fbsacap@leeds.ac.uk

Phone: +44(0) 113 343 4645

ABSTRACT

A subthreshold pulse of transcranial magnetic stimulation (TMS) on the motor cortex can modulate the amplitude of the monosynaptic reflex (H-reflex) elicited in the flexor carpi radialis (FCR) muscle, a method known as TMS-conditioning of the H-reflex. The purpose of this study was to establish the intersession reliability of this method over the course of three sessions. Eleven healthy participants received either peripheral nerve stimulation (PNS), TMS or a combination of the two. The intensity of the PNS stimuli was set to evoke a monosynaptic response (H-reflex) corresponding to 10% of the maximum motor response (M_{max}), $H_{M10\%}$. The conditioning effect of TMS on the monosynaptic reflex was assessed by delivering subthreshold cortical pulses at different conditioning-test intervals (from -7 ms to 7 ms) from peripheral nerve stimulation. The first interval at which facilitation could be observed was deemed early facilitation (EF). Using intraclass correlation coefficients (ICCs), we found excellent reliability for M_{max} amplitudes (ICC = 0.98), $H_{M10\%}$ amplitudes (ICC = 0.85) and TMS-conditioned H-reflexes recorded at the interval following EF (EF+2 ms) (ICC = 0.87). Good reliability (ICCs ranging from 0.67 to 0.77) was found for the other conditioning-test intervals. We conclude that TMS-conditioned H-reflexes are reliable parameters to assess the excitability of corticospinal circuits.

Keywords: Transcranial magnetic stimulation, H-reflex, FCR, corticospinal tract, reliability

Abbreviations: EF, early facilitation; EMG, electromyography; FCR, flexor carpi radialis; H-reflex, Hoffman reflex; M_{max}, maximal motor wave; $H_{M10\%}$, H-reflex at 10% of M_{max}; ICC, intraclass correlation coefficient; MEP, motor evoked potential; M1, primary motor cortex; MSO, maximum stimulator output; MT, motor threshold; PNS, peripheral nerve stimulation; TMS, transcranial magnetic stimulation.

1. Introduction

The excitability of corticospinal circuits can be assessed non-invasively in humans using different techniques. Transcranial magnetic stimulation (TMS) of the primary motor cortex (M1) induces a motor evoked potential (MEP) in the muscle recorded via surface electromyography (EMG) [1]. The MEP amplitude can be used to estimate the excitability of the corticospinal tract, but depends on motoneuron excitability [2]. Electrical stimulation of a peripheral nerve (peripheral nerve stimulation, PNS) produces two key responses in the recorded EMG. First, stimulation of the motor nerve fibers produces a short-latency response named motor wave (M-wave), which reflects the excitability of motor axons [3]. Second, stimulation of the sensory fibers produces a monosynaptic reflex (H-reflex). Transcranial Magnetic Stimulation can also be used to evaluate descending influences on spinal reflex excitability through modulation of the H-reflex, or TMS-conditioning of the monosynaptic reflex [4]. Specifically, participants are stimulated at rest as muscle activity of the FCR is recorded and subthreshold TMS and median nerve stimulation at different time intervals are delivered. This causes an increase in the amplitude of the H-reflex when elicited before (2 ms) and after (up to 4 ms) the cortical stimulus. The early increase corresponds to the direct (pyramidal) volley to spinal motoneurons, while the late increase is due to polysynaptic pathways to spinal motoneurons [5].

Recently, conditioning of the H reflex has been employed to differentiate between the direct and indirect waves generated by cortical stimulation, representing multiple descending volleys to spinal motoneurons [6]. In order to do so, PNS and TMS were delivered in combination at conditioning-test intervals ranging from -7 (PNS first) to 8 (TMS first) ms. The first interval at which the amplitude of the resulting conditioned H-reflex increased compared to the unconditioned H-reflex amplitude was termed early facilitation (EF). Thus it is assumed that data collected at the EF interval represent the direct (e.g. monosynaptic) component of the cortical descending volley, while data collected at later intervals (e.g. EF+2, EF+4) represent instead increasingly longer polysynaptic pathways to spinal motoneurons [7].

When assessing changes in central nervous system excitability occurring after a CNS injury or motor training, it is crucial to employ validated and reliable techniques [8]. An example of such a validated technique is the estimation of the TMS motor threshold (MT). The MT is defined as the minimum stimulation intensity necessary to evoke MEPs in the muscle of interest [9] and the intensity of the cortical pulse used to condition the H-reflex is based on its value. MTs exhibit excellent (ICC = 0.97) inter-session reliability when stimulating the FCR muscle [10]. In addition, the monosynaptic reflex evoked in the FCR muscle via stimulation of the median nerve was also noted to have high degree of intersession reliability (ICC = 0.92) over four separate days [11]. When recorded from the soleus muscle, moderate-to-good reliability was observed for the short-latency (ICC = 0.71) and long-latency (ICC = 0.45) facilitation of the monosynaptic reflex upon TMS-conditioning [8]. Nevertheless, to the best of our knowledge, the intersession reliability of the TMS-conditioned H-reflex in forearm muscles is yet to be established. The aim of the current project was to examine how reliable forearm muscle responses obtained upon cortical and spinal stimulation are over three sessions by measuring the intraclass reliability of: (A) maximal motor wave amplitude (M_{max}); (B) H-reflex amplitude to a stimulus at 10% of M_{max} ($H_{M10\%}$); (C) TMS-conditioned monosynaptic reflex (TMS-PNS) at multiple conditioning-test intervals.

2. Materials and Methods

2.1. Participants

Thirteen participants (mean age = 26.07, SD = 3.69, females = 6) volunteered for the study. Consecutive testing sessions were separated by at least 24 hours in order to avoid any carryover effects induced by the TMS protocol, and at the same time of day to control for any potential influence of circadian rhythms [12]. All participants gave written informed consent to procedures approved by the Faculty of Biological Sciences Ethical Review Committee at the University of Leeds and in accordance with the declaration of Helsinki. Participants were included in the study only if the H-reflexes recorded from their right FCR did not overlap with the motor waves recorded from the same muscle, rendering the interpretation of the recording difficult: two participants out of the original thirteen were excluded from the study for this reason ($n = 11$).

2.2. Recording techniques

Participants were seated with hip and knees forming an angle of 90°, feet resting on foot support, the right forearm in full pronation and the elbow flexed at an angle of 120° supported by a dynamometer (Biodex Corp., Shirley, NY). Electromyography activity was recorded from the right flexor carpi radialis (FCR) by means of a parallel-bar wireless mini sensor (2.5 x 1.2 cm) (Trigno, Delsys Inc., Natick, MA, USA). The optimal location to record activity from the FCR muscle is reported to be at one third of the distance between the medial epicondyle and the radial styloid [11]. Markings and pictures of the position of the electrodes were taken on each session to ensure the consistency of recordings across days. The EMG signal was pre-amplified (gain = 909), recorded with a 20-450 Hz bandwidth and digitized at 2 kHz using data acquisition software (Spike2, Cambridge electronics Design, Cambridge, UK). All measurements were performed at rest.

2.3. Stimulation techniques

Magnetic stimuli were delivered to the left M1 by a Magstim Rapid stimulator with the coil (70mm Double Air Firm coil, Magstim Company, Whitland, Dyfed, UK) being held by a support stand (Magstim AFC Support Stand, Magstim Company, Whitland, Dyfed, UK), oriented at ~45° to the sagittal plane to induce a posterior-to-anterior current flow across the motor strip [13]. The optimal coil position to evoke MEPs in FCR was found by moving the coil over the scalp while delivering stimulation and by marking the position at which MEPs could be elicited with the lowest stimulation intensity. In order to ensure the consistency of recordings across sessions, we acquired pictures of the coil position and orientation and measured the distance from the vertex to the stimulation site. During all the interventions, the stimulation was controlled through Spike2 software (Cambridge Electronic Design, Cambridge, UK).

Peripheral nerve stimulation (PNS) targeted the median nerve at the forearm. The most reliable locus of stimulation for eliciting activity in FCR is the cubital fossa, medial to the tendon of biceps brachii, parallel to the nerve [14]. We used a bar stimulating electrode (E.SB010, Digitimer Ltd, Welwyn Garden City, UK) consisting of anode and cathode stainless steel electrodes of 8 mm diameter and spaced 30 mm apart. The stimulation was delivered through a constant-current stimulator (DS7A, Digitimer Ltd, Welwyn Garden City, UK) which was controlled by the acquisition software (Spike2, Cambridge Electronic Design, Cambridge, UK). The median nerve was stimulated using 1 ms monophasic pulses [2].

2.4. Experimental procedure

The recording procedure started with either TMS or PNS alone, in a randomized order. TMS – PNS conditioning was always delivered last as the stimulation parameters used during this phase of the session are derived from the outcomes of the TMS and PNS phases alone. EMG activity was recorded continuously during the experiment to ensure lack of changes from baseline.

PNS alone

Electrical stimulation started at low intensity (1.0 mA) and was then increased in steps of 0.2 mA until a monosynaptic reflex was discernible from the EMG recordings at ~15-20 ms after median nerve stimulation [3, 15]. To estimate the maximum motor response (M_{max}), the intensity of the stimulator output was incremented in steps of 0.3 mA starting from the intensity at which a monosynaptic reflex could first be evoked until the peak-to peak amplitude of the M-wave reached its plateau. Ten traces were obtained at the intensity of stimulation at which the M-wave was maximal.

TMS alone

Once the optimal location on the scalp to induce activity in FCR was located, markings were made directly on the scalp to aid guidance of the coil. For each participant and in each session, an individual resting motor threshold (MT) was estimated. The MT is defined as the lowest TMS intensity, given as a percentage of the maximum stimulator output (MSO), which elicits MEPs with peak-to-peak amplitudes of $>50\ \mu$V in at least 5 out of 10 traces [9]. The interval between two pulses was set at 5 seconds.

TMS – PNS conditioning tests

We measured the effects of delivering cortical stimulation at different conditioning-test intervals from the peripheral nerve stimulation ranging from -7 ms (PNS first) to 7 ms (TMS first) (Fig. 1). We chose this range of intervals based on previous studies assessing a conditioned H-reflex in the FCR muscle [6]. The conditioning TMS pulse was given at 90%MT intensity, an intensity which should not elicit electrical activity in the recorded EMG but can elicit descending activity along the corticospinal tract and modulate the excitability of the spinal MN pool [6]. The intensity of the electrical stimulation was set to produce H-reflex amplitudes equivalent to 10% of M_{max} ($H_{M10\%}$) [3]. This intensity is chosen so that the H-reflex is elicited in the ascending part of its recruitment curve and is not contaminated by antidromic propagation of motor axons action potentials [16]. For each conditioning-test interval, the mean amplitude of the 8 conditioned reflexes was normalised to the mean unconditioned H-reflex. Therefore, data collected at each conditioning-test interval were quantified as a percentage of the unconditioned H-reflex [3]. We obtained 8 EMG traces for the unconditioned H-reflex.

2.5. Data analyses

The conditioning-test interval at which the peripheral afferent volley and the monosynaptic component of the corticospinal volley reach spinal motoneurons simultaneously (early facilitation, EF) was estimated as the first interval for which conditioned H-reflex amplitudes increased from the unconditioned amplitude values [6]. Changes from baseline were assessed via uncorrected paired Student's tests between the unconditioned amplitudes and the conditioned values at each interval. This procedure was completed for each participant and in each session. Conditioning test-intervals following EF were aligned to it (e.g. EF+2) across participants and sessions.

Statistical analyses were performed using SPSS (Version 22.0) software with an *a priori* alpha level of .05. A linear mixed-effects model fit by maximum likelihood was specified to assess the effects of Interval (HM10%, EF. EF+2, EF+4, EF+6), Session (1,2,3) and their interaction in explaining variations in Amplitude. By-subject random intercepts and random slopes were added to account for the non-independency of observations, with a variance components covariance structure. We tested three alternative models including the following fixed effects: one with Interval, Session and the interaction effect; one without the interaction effect; one without the interaction effect and the main effect of Session. Likelihood ratio tests were carried out to assess which model better fitted the data. The distributions of residuals were plotted to check for any violation of the assumption of normality. Differences between conditioned and unconditioned reflexes obtained during the three sessions were assessed using post-hoc tests and results from multiple comparisons were corrected with the Bonferroni procedure. We report means and 95% confidence intervals (CIs) for all the parameters.

The intraclass reliability is a measure of consistency between measurements, and its value (the intraclass correlation coefficient, ICC) ranges from 0 to 1 with 1 indicating perfect similarity [17]. A 2-ways mixed effects model was used to calculate ICCs following the equation given by Koo and Li [17]. Reliability analyses were performed for the following parameters: M_{max}, $H_{M10\%}$, EF, EF+2, EF+4, EF+6. ICCs values were interpreted as follow: 0.81 to 1, excellent; 0.61 to 0.80, good; 0.41 to 0.60, moderate; 0.21 to 0.40, fair; below 0.20, poor [18].

3. Results

3.1. TMS-conditioned monosynaptic reflexes

A reliable H-reflex could be obtained at rest in 85% (11/13) of the participants originally recruited. Mean values, standard deviations and range of values for all the recordings are reported in Table 1. The results from likelihood-ratio tests of goodness-of-fit revealed that the model including only the fixed effect of Interval (*Log L* = 143.320, *AIC* = 159.320, *N of parameters* = 8) provided a better fit of the dataset than the model including Session as a fixed effect (*Log L* = 141.426, *AIC* = 161.231, *N of parameters* = 10) and the model including Session and the interaction between Interval and Session as fixed effects (*Log L* = 138.533, *AIC* = 174.553, *N of parameters* = 18). The plotted distribution of residuals did not show substantial variations from normality. The fixed factor Interval affected the amplitude of the recorded values (F = 12.226, $P < 0.001$). Fixed Effects estimates are reported in Table 2 alongside their *p* values and 95% CIs. Post-hoc analyses revealed that TMS increased the size of the H-reflex at all intervals from the early facilitation: EF ($P < 0.001$), EF+2 ($P < 0.001$), EF+4 ($P < 0.001$), and EF+6 ($P < 0.001$). Individual and mean values, expressed as percentages of the control unconditioned H-reflex, for each session are presented in Fig. 2.

3.2. Reliability analysis

Results from the ICCs analysis are reported in Table 3. Excellent between-session reliability was observed for $M_{max,}$ (ICC = 0.98, F = 274.95, P <0.001), $H_{M10\%}$ (ICC = 0.85, F = 17.70, P <0.001) and EF+2 (ICC = 0.87, F = 21.68, P <0.001) (Fig. 3). Good reliability was found for EF (ICC = 0.77, F = 11.23, P <0.001), EF+4 (ICC = 0.72 F = 8.61, P <0.001) and EF+6 (ICC 0.67, F = 7.02, P <0.001).

4. Discussion

The main aim of the current project was to determine the intersession reliability of a series of neurophysiological parameters recorded from forearm muscles (FCR), which may be useful to characterise changes in the excitability of corticospinal circuits occurring after lesions or motor training. We replicated the finding [4, 5] that TMS increases the amplitude of the monosynaptic reflex evoked from FCR when given at a range of conditioning-test intervals from the peripheral pulse (Fig. 2). Furthermore, we showed that the intersession reliability of this phenomenon varied when using different conditioning-test intervals, with the highest degree of reliability obtained at the interval for which the early cortical volley reached spinal motoneurons 2 ms before the afferent volley (Table 3). Thus, the main findings of the study indicate that TMS-conditioning of the H-reflex evoked in the FCR muscle is a reliable technique to measure pathway-specific plasticity.

4.1. TMS-conditioned monosynaptic reflexes

Cortical stimulation modulated the amplitude of the monosynaptic reflex when the descending volley induced by the subthreshold pulse reached the spinal cord up to 6 ms before the afferent volley. The observed mean peaks of facilitation in the current study were 207% on the first session, 217% on the second session and 221% in the third session. Mazzocchio and colleagues [5] employed a similar experimental setting as described in the current study (use of a subthreshold TMS pulse and parameters recorded at rest) and reported facilitation of the evoked response by up to 130% of the unconditioned values. A potential explanation for the higher degree of facilitation observed in our investigation is the use of different stimulation parameters. We chose to stimulate the median nerve at an intensity which elicits a relatively small reflex (10% of M_{max}) in the muscle of interest to preferentially activate low-threshold Ia fibres and minimize the potential collision with the motor wave [16]. Moreover, we employed a coil orientation (posterolateral to anteromedial) inducing an electrical field perpendicular to the central sulcus and leading to larger MEPs [16]. More recent studies [6] reported mean facilitation values as high as 200% with this coil orientation.

Multiple hypotheses have been advanced on the nature of the facilitation observed when the timing between peripheral and cortical stimulation is manipulated. Based on the difference in latencies between the recorded MEP and H-reflexes, Cowan and his colleagues [19] first argued that the first facilitation occurred because of a synchronous activation of spinal motoneurons brought upon by the two forms of stimulation. This facilitation is likely to be mediated by large-diameter corticospinal axons, which synapse directly on spinal motoneurons. This interval would correspond to the early facilitation (EF) observed in this study. Different neural populations could be involved in the later stages of facilitation. Conditioning effects occurring at the EF+2 interval could be due to the late arrival of slow-conducting corticospinal tract neurons at the spinal motoneuron pools [19], or represent disynaptic routes to spinal motoneurons via spinal interneurons [20]. For example, interneurons mediating presynaptic inhibition of Ia afferents can be activated by cortical stimulation [21]. Spinal motoneurons receive monosynaptic and polysynaptic inputs from other pathways like the reticulospinal, rubrospinal and vestibulospinal tracts, all of which may alter the excitability of spinal circuits and influence the amplitude of the monosynaptic reflex [22]. Finally, it has recently been demonstrated [6] that the time course of TMS-conditioning of the FCR H-reflex closely matches the late descending volleys (I waves) induced by TMS, that is thought to originate from M1 interneuron circuits [23].

4.2. Reliability analysis

The maximal amplitude of the motor wave (M_{max}) evoked in response to electrical stimulation of a peripheral nerve is elicited by recruitment of all motor axons [3]. Monitoring changes in M_{max} is important to ensure that there were no changes in participants' position, location of the stimulating and recording electrodes or any muscular effects which may influence the recordings. This parameter showed excellent intersession reliability, in line with previous studies [11]. Difficulties in reporting a monosynaptic reflex in the FCR muscle at rest have been previously reported [24]. A widely used method to increase excitability and facilitate the occurrence of a reflex is to ask participants to contract the muscle slightly while receiving stimulation [16] However, since the effects of TMS on the monosynaptic reflex differ during flexion [4], we decided to exclude two participants from our pool of thirteen because a reliable reflex could not be evoked or was cancelled due to collision with the motor wave at rest, rather than asking them to contract the muscle. The intensity of the peripheral stimulus was chosen to evoke a reflex response in the ascending portion of the recruitment curve, at which the influence of Ib afferents is lower [16]. We found that this parameter was highly stable across sessions (see Table 3).

We conditioned the H-reflex with TMS, delivered at different intervals relative to the peripheral stimulus. The ICC was highest (0.87) at EF+2 ms. At this interval, the corticospinal volley may have reached spinal motoneurons slightly before the arrival of the afferent volley. The interval corresponding to the direct corticomotoneuronal transmission (EF) was less reliable (ICC = 0.77). This indicates that the subthreshold cortical pulse did not excite spinal motoneurons through corticomotoneuronal connections always to the same extent [25]. Both correlation coefficients are higher than the one observed when TMS precedes electrical stimulation of the soleus muscles [8]. However, the response latencies of MEPs and H-reflexes evoked in the soleus muscle differ from the ones obtained in FCR and a comparison between the reported ICCs is not straightforward. Importantly, in the current study, we tested a range of intervals while Gray and his colleagues only investigated long-latency effects (10 ms interval). Reliability was lower for the two intervals following EF+2. A possible explanation for this decrease may be the polysynaptic nature of the conditioned responses. With longer times between the two stimuli, the chance of indirect descending pathways to influence the reflex increases and so does the variability of responses [26].

The importance of studies using TMS to assess motor excitability is hindered by the high variability of results and lack of reproducibility [27]. A series of issues need to be considered when considering the outcomes of cortical stimulation [2]. The possibility that confounding effects not directly related to the stimulation protocol such as participants' attention and muscle pre-activation may affect the responses we collected cannot be excluded. Nonetheless, the cortical pulse increased only the size of the H-reflexes with no effects on the M response, as would be expected if the changes occurred because of differences in muscle pre-activation or attention [28]. In addition, changes in the activation state of the participant [4] were controlled for by monitoring any variation in the EMG recorded from FCR occurring immediately prior to any stimulation. A possible limitation of the study is the relatively small sample of participants. This is, however, in line with previous studies assessing intersession reliability of the H-reflex [8, 29]. Moreover, reliability studies are often limited to measure the stability of parameters over 2 consecutive sessions. As clinical practices and rehabilitation protocols may require a higher number of sessions to be implemented [8], we chose to include a third session in the current study.

5. Conclusions

Taken together, our findings indicate that the conditioning effect of TMS on the monosynaptic reflex evoked in FCR muscle is a reliable phenomenon. Its intersession reliability is higher at the conditioning-test interval for which the cortical volley reaches spinal motoneurons 2 ms before the afferent volley (EF +2). TMS-conditioning of the H-reflex can be used to study pathway-specific changes in neural excitability underlying motor control and motor learning.

Declarations of interest: none.

Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

REFERENCES

[1] M. Hallett, Transcranial magnetic stimulation: a primer, Neuron 55 (2007) 187-199.
[2] D. Burke, E. Pierrot-Deseilligny, Caveats when studying motor cortex excitability and the cortical control of movement using transcranial magnetic stimulation, Clinical Neurophysiology 121 (2010) 121-123.
[3] R.M. Palmieri, C.D. Ingersoll, M.A. Hoffman, The Hoffmann reflex: methodologic considerations and applications for use in sports medicine and athletic training research, Journal of Athletic Training 39 (2004) 268.

[4] J. Nielsen, N. Petersen, G. Deuschl, M. Ballegaard, Task-related changes in the effect of magnetic brain stimulation on spinal neurones in man, The Journal of Physiology 471 (1993) 223-243.

[5] R. Mazzocchio, J. Rothwell, B. Day, P. Thompson, Effect of tonic voluntary activity on the excitability of human motor cortex, The Journal of Physiology 474 (1994) 261-267.

[6] N. Niemann, P. Wiegel, A. Kurz, J.C. Rothwell, C. Leukel, Assessing TMS-induced D and I waves with spinal H-reflexes, Journal of Neurophysiology 119 (2017) 933-943.

[7] C. Leukel, W. Taube, J. Rittweger, A. Gollhofer, M. Ducos, T. Weber, J. Lundbye-Jensen, Changes in corticospinal transmission following 8 weeks of ankle joint immobilization, Clinical Neurophysiology 126 (2015) 131-139.

[8] W. Gray, M. Sabatier, T. Kesar, M. Borich, Establishing between-session reliability of TMS-conditioned soleus H-reflexes, Neuroscience Letters 640 (2017) 47-52.

[9] P.M. Rossini, A. Barker, A. Berardelli, M. Caramia, G. Caruso, R. Cracco, M. Dimitrijević, M. Hallett, Y. Katayama, C. Lücking, Non-invasive electrical and magnetic stimulation of the brain, spinal cord and roots: basic principles and procedures for routine clinical application. Report of an IFCN committee, Electroencephalography and Clinical Neurophysiology 91 (1994) 79-92.

[10] M. Malcolm, W. Triggs, K. Light, O. Shechtman, G. Khandekar, L.G. Rothi, Reliability of motor cortex transcranial magnetic stimulation in four muscle representations, Clinical Neurophysiology 117 (2006) 1037-1046.

[11] A.D. Christie, J.G. Inglis, J.P. Boucher, D.A. Gabriel, Reliability of the FCR H-reflex, Journal of Clinical Neurophysiology 22 (2005) 204-209.

[12] M.V. Sale, M.C. Ridding, M.A. Nordstrom, Factors influencing the magnitude and reproducibility of corticomotor excitability changes induced by paired associative stimulation, Experimental Brain Research 181 (2007) 615-626.

[13] J. Rothwell, Techniques and mechanisms of action of transcranial stimulation of the human motor cortex, Journal of Neuroscience Methods 74 (1997) 113-122.

[14] S. Jaberzadeh, S. Scutter, A. Warden-Flood, H. Nazeran, Between-days reliability of H-reflexes in human flexor carpi radialis, Archives of Physical Medicine and Rehabilitation 85 (2004) 1168-1173.

[15] M. Hugon, Methodology of the Hoffmann reflex in man. Human Reflexes, Pathophysiology of Motor Systems, Methodology of Human Reflexes, Vol. 3, Karger Publishers, 1973, pp. 277-293.

[16] E. Pierrot-Deseilligny, D. Mazevet, The monosynaptic reflex: a tool to investigate motor control in humans. Interest and limits, Neurophysiologie Clinique/Clinical Neurophysiology 30 (2000) 67-80.

[17] T.K. Koo, M.Y. Li, A guideline of selecting and reporting intraclass correlation coefficients for reliability research, Journal of Chiropractic Medicine 15 (2016) 155-163.

[18] D.G. Altman, Practical statistics for medical research, London: Chapman and Hall, 1991.

[19] J. Cowan, B. Day, C. Marsden, J. Rothwell, The effect of percutaneous motor cortex stimulation on H reflexes in muscles of the arm and leg in intact man, The Journal of Physiology 377 (1986) 333-347.

[20] E. Jankowska, Y. Padel, R. Tanaka, Disynaptic inhibition of spinal motoneurones from the motor cortex in the monkey, The Journal of Physiology 258 (1976) 467-487.

[21] S. Meunier, E. Pierrot-Deseilligny, Cortical control of presynaptic inhibition of Ia afferents in humans, Experimental Brain Research 119 (1998) 415-426.

[22] R.M. Eccles, A. Lundberg, Supraspinal control of interneurones mediating spinal reflexes, The Journal of Physiology 147 (1959) 565-584.

[23] V. Di Lazzaro, P. Profice, F. Ranieri, F. Capone, M. Dileone, A. Oliviero, F. Pilato, I-wave origin and modulation, Brain Stimulation 5 (2012) 512-525.

[24] T. Miller, A. Newall, D. Jackson, H-reflexes in the upper extremity and the effects of voluntary contraction, Electromyography and Clinical Neurophysiology 35 (1995) 121-128.

[25] N. Niemann, P. Wiegel, J.C. Rothwell, C. Leukel, The effect of subthreshold transcranial magnetic stimulation on the excitation of corticospinal volleys with different conduction times, bioRxiv (2016) 084574.

[26] C. Ribrault, K. Sekimoto, A. Triller, From the stochasticity of molecular processes to the variability of synaptic transmission, Nature Reviews Neuroscience 12 (2011) 375.

[27] M.E. Héroux, J.L. Taylor, S.C. Gandevia, The use and abuse of transcranial magnetic stimulation to modulate corticospinal excitability in humans, PLoS One 10 (2015) e0144151.

[28] M. Knikou, The H-reflex as a probe: pathways and pitfalls, Journal of Neuroscience Methods 171 (2008) 1-12.

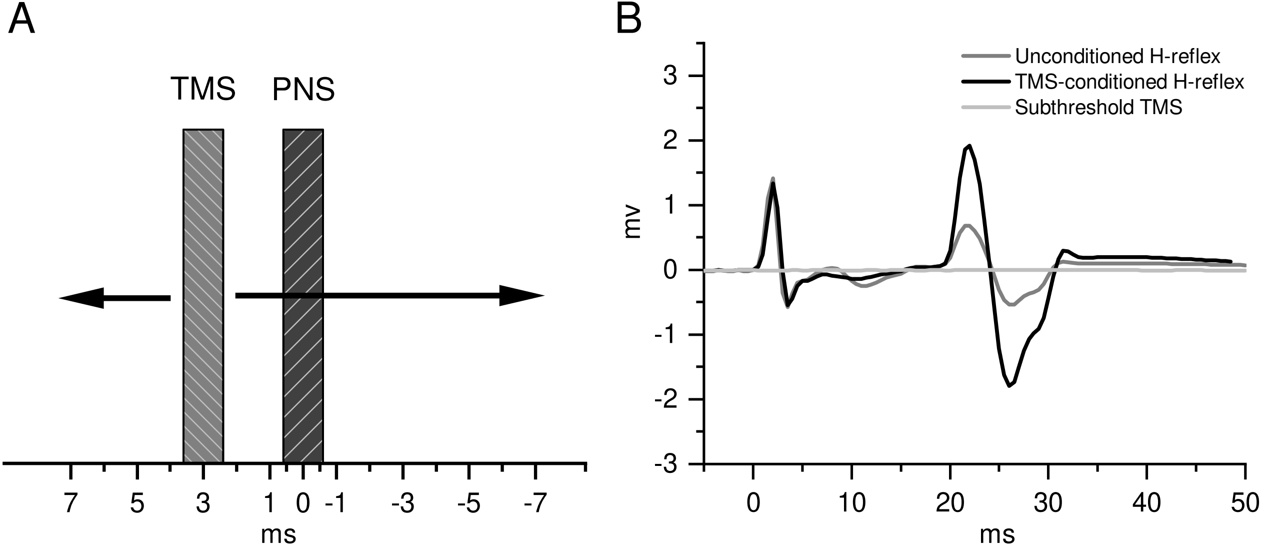
[29] M.C. Hoch, B.A. Krause, Intersession reliability of H: M ratio is greater than the H-reflex at a percentage of M-max, International Journal of Neuroscience 119 (2009) 345-352.

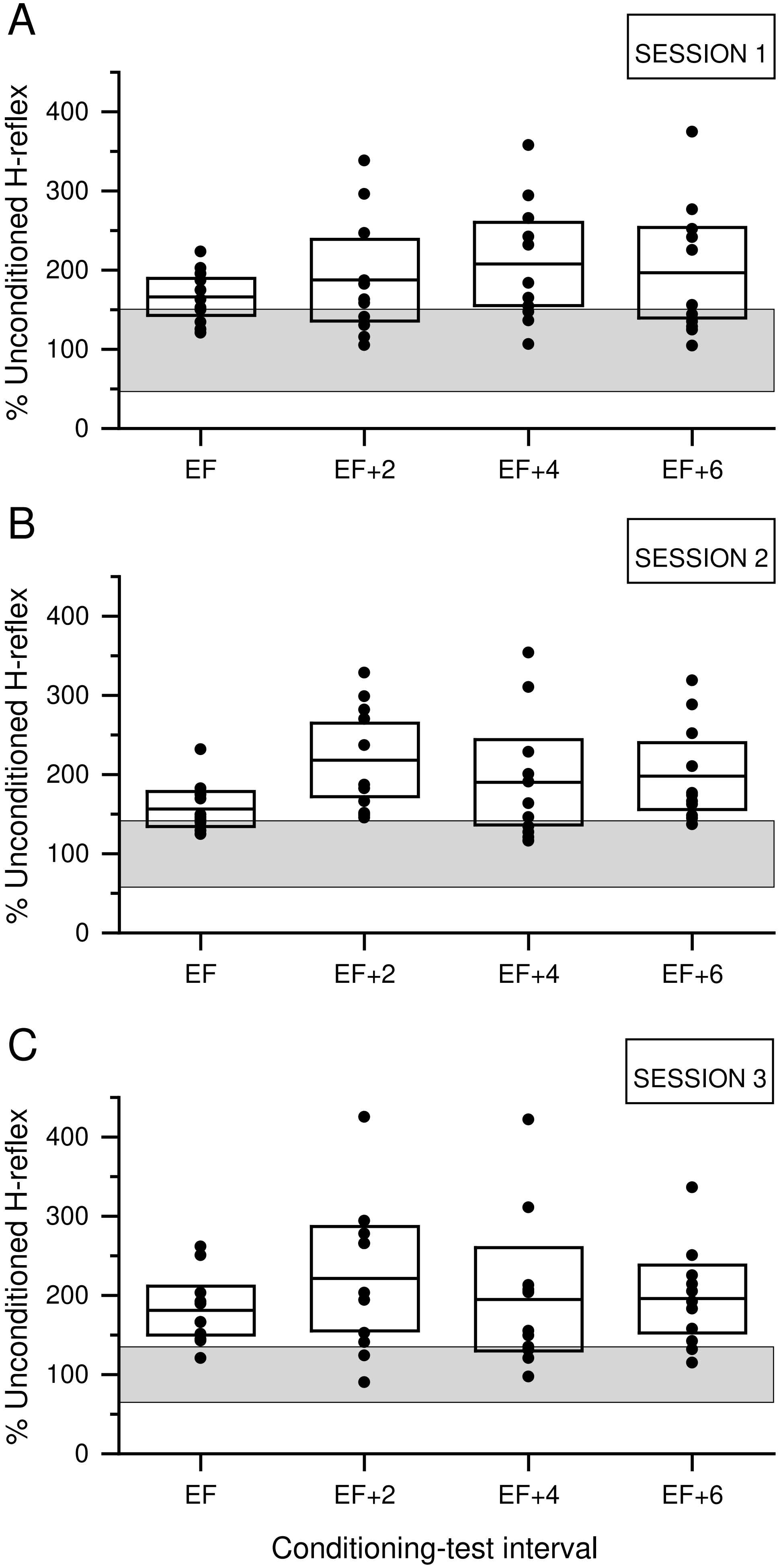
Figure legends

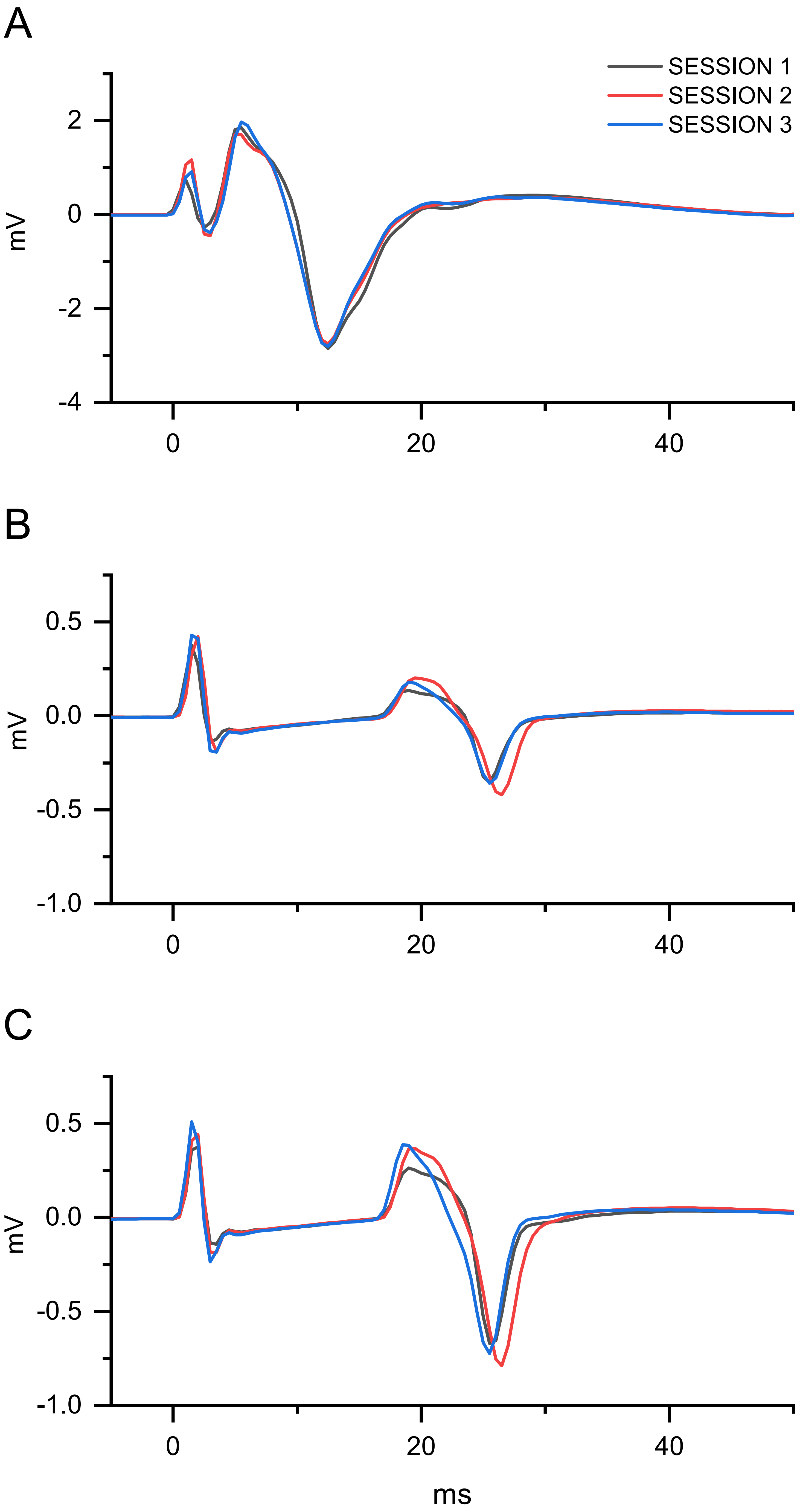
Fig.1. TMS-conditioned monosynaptic reflex protocol. (A) TMS was delivered at ISIs ranging from -7 to 7 ms (3 ms in the example). A negative conditioning-test interval indicates that the conditioning stimulus (TMS) was applied after the test stimulus (PNS). (B) TMS-conditioned H-reflex amplitude (3 ms conditioning-test interval) compared to the unconditioned H-reflex in a representative participant. TMS alone (Subthreshold MT) did not produce any MEP.

Fig. 2. TMS-conditioned H-reflexes. Individual (*n* = 11) means of the conditioned H-reflexes, given as percentages of the unconditioned H-reflex mean, at different conditioning-test intervals for SESSION 1 (A), SESSION 2 (B) and SESSION 3 (C). The top and bottom lines of the boxes represent the associated 95% CIs. The black lines represent the means. The grey shaded areas represent the 95% CIs of the control unconditioned H-reflex value.

Fig. 3. Intersession reliability of the recordings in a representative participant. (A) Stability of the Mmax over 3 sessions. (B) Stability of the unconditioned H-reflex over 3 sessions. (C) Stability of the TMS-conditioned H-reflex at the EF interval over 3 sessions. Each trace represents the mean of 8 sweeps.


A
TMS
PNS
7
5
3
1
0
-1
-3
-5
-7
ms
B
Unconditioned H-reflex
TMS-conditioned H-reflex
Subthreshold TMS
mv
3
2
1
0
-1
-2
-3
0
10
20
30
40
50
ms


A
SESSION 1
% Unconditioned H-reflex
0
100
200
300
400
EF
EF+2
EF+4
EF+6
B
SESSION 2
% Unconditioned H-reflex
0
100
200
300
400
EF
EF+2
EF+4
EF+6
C
SESSION 3
% Unconditioned H-reflex
0
100
200
300
400
EF
EF+2
EF+4
EF+6
Conditioning-test interval


A
SESSION 1
SESSION 2
SESSION 3
2
0
-2
-4
mV
0
20
40
B
0.5
0.0
-0.5
-1.0
mV
0
20
40
C
0.5
0.0
-0.5
-1.0
mV
0
20
40
ms