A systematic method to quantify the presence of cross-talk in stimulus-evoked EMG responses: Implications for TMS studies

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Selvanayagam VS, Riek S, Carroll TJ. A systematic method to quantify the presence of cross-talk in stimulus-evoked EMG responses: Implications for TMS studies. J Appl Physiol 112: 259–265, 2012. First published November 3, 2011; doi:10.1152/japplphysiol.00558.2011.—Surface electromyography (EMG) responses to noninvasive nerve and brain stimulation are routinely used to provide insight into neural function in humans. However, this could lead to erroneous conclusions if evoked EMG responses contain significant contributions from neighboring muscles (i.e., due to “cross-talk”). We addressed this issue with a simple nerve stimulation method to provide quantitative information regarding the size of EMG cross-talk between muscles of the forearm and hand. Peak to peak amplitude of EMG responses to electrical stimulation of the radial, median, and ulnar nerves (i.e., M-waves) were plotted against stimulation intensity for four wrist muscles and two hand muscles (n = 12). Since electrical stimulation can selectively activate specific groups of muscles, the method can differentiate between evoked EMG arising from target muscles and EMG cross-talk arising from nontarget muscles. Intramuscular EMG responses to nerve stimulation and root mean square EMG produced during maximal voluntary contractions (MVC) of the wrist were recorded for comparison. Cross-talk was present in evoked surface EMG responses recorded from all nontarget wrist (5.05–39.38% Mmax) and hand muscles (1.50–24.25% Mmax) and to a lesser degree in intramuscular EMG signals (~3.7% Mmax). The degree of cross-talk was comparable for stimulus-evoked responses and voluntary activity recorded during MVC. Since cross-talk can make a considerable contribution to EMG responses in forearm and hand muscles, care is required to avoid misinterpretation of EMG data. The multiple nerve stimulation method described here can be used to quantify the potential contribution of EMG cross-talk in transcranial magnetic stimulation and reflex studies.

surface electromyography; nerve stimulation; reflex; biomechanics

IN RECENT YEARS, MAGNETIC and electrical stimulation techniques have been applied in many contexts to better understand human motor control. These noninvasive stimulation techniques are generally applied at the level of the motor cortex (1, 5, 14), cervicomedullary junction (13, 22), or peripheral nerves (i.e., for reflex studies; e.g., Ref. 12) to provide information about neural excitability. The responses produced by these stimulation techniques are commonly recorded at the target muscle using surface electromyography (sEMG), which gives global information about muscle activation, is noninvasive, and convenient to apply. However, in using sEMG, there is a risk of recording “cross-talk” (3), which occurs when signals are recorded from nontarget muscles that are in close proximity to the muscle of interest. Undetected cross-talk can lead to serious misinterpretations of stimulus-evoked sEMG data. For example, Myklebust et al. (16) proposed the existence of a “reciprocal excitatory reflex” on the basis of responses detected from electrodes over the anterior tibialis muscle in response to stimulation of the antagonist tibial nerve. Subsequent studies showed that the supposed anterior tibialis response was actually the result of cross-talk from the soleus muscle (8, 21). Therefore, the identification of a convenient method that is applicable under standard transcranial magnetic stimulation (TMS) and reflex study conditions to differentiate between sEMG signals arising from cross-talk from those generated in the target muscle would represent a critical technical advance.

Methods aimed at quantifying or reducing cross-talk have been proposed previously (2, 7, 9, 11, 23, 24), but perhaps because they were framed in the context of measurement of voluntary muscle activity, they are not commonly considered during studies involving noninvasive stimulation techniques such as TMS. There has been a rapid increase in the number of TMS studies reported in the past 5–10 years, and the majority of these studies has focused on muscles of the upper limb (i.e., forearm and hand muscles). While the presence of sEMG cross-talk arising from electrical nerve stimulation between muscles of the lower limb has been identified (3, 21, 23), we are not aware of studies that have systematically documented the presence of stimulus-evoked sEMG cross-talk among muscles of the upper limb. Since the forearm and hand muscles are grouped closely together, we expect that sEMG cross-talk arising from electrical stimulation might be much higher than those found in the lower limb (15, 24). Thus there is a need to both highlight the potential risks of cross-talk in hand and forearm muscles and identify a simple and convenient method to ensure that inadvertent recording of signals from nontarget muscles does not lead to incorrect interpretation of TMS studies.

Here we describe a systematic method of quantifying the degree of sEMG cross-talk between forearm [flexor carpi radialis (FCR), flexor carpi ulnaris (FCU), extensor carpi radialis brevis (ECRb), and extensor carpi ulnaris (ECU)] and hand muscles [abductor pollicis brevis (APB) and adductor digiti minimi (ADM)] by electrically stimulating multiple nerves proximal to the elbow (radial, median, and ulnar nerves). Since electrical nerve stimulation can selectively activate specific groups of target muscles, this approach allows EMG signals recorded from both target (M-wave) and nontarget (cross-talk) muscles to be quantified. The method can be applied in 5–10 min during standard recording conditions common in TMS or reflex studies. We also made comparisons between the amount of the stimulus-evoked cross-talk recorded from surface vs. intramuscular (fine wire) electrodes and between the degree of cross-talk apparent in response to stimulus-
evoked responses vs. sEMG produced by voluntary contraction at the wrist. The study tested the hypotheses that sEMG cross-talk between muscles of both the forearm and hand can be large and that the problem of cross-talk is greater for stimulus-evoked EMG responses than for voluntary contractions. Confirmation of this hypothesis would have serious implications for the interpretation of many TMS and reflex studies.

MATERIALS AND METHODS

Participants. Sixteen healthy volunteers (14 men, 2 women) without a history of neurological disease and with a mean age of 28.1 ± 6.5 yr were recruited to participate in the study. Data from four volunteers that contained evidence of cross-nerve stimulation were removed from analysis. Cross-nerve stimulation was revealed by an increase in recruitment gradient in EMG signals of nontarget muscles (i.e., FCU). This increase occurred well after the recruitment threshold for the target muscle in response to the increasing stimulation intensity of the noninnervating nerve (i.e., median nerve). In addition, three volunteers participated in an additional study using fine wire EMG. All participants provided their written informed consent to the procedures and protocols of the study, which conform to the Declaration of Helsinki and are approved by the University of Queensland Ethics Committee.

Setup and general procedure. The setup of the experiment was similar to that described by Lee and Carroll (10), de Rugy and Carroll (4), and Selvanayagam et al. (20). The participants were seated on a comfortable chair with the right arm snug in a custom-made device in a neutral forearm position (i.e., midpoint between supination and pronation) with the elbow joint at ∼110° angle. The hand at the metacarpo-phalangeal joint and the wrist joint were padded with a polyurethane foam strap (0.3 cm thick) and silicone rubber foam strap (0.6 cm thick), respectively, and secured by adjustable surface clamps on all four sides of the joints. The device measured radial and ulnar deviation force of the wrist joint in the vertical plane and a flexion and extension force in the horizontal plane in response to both voluntary contractions and electrical stimulation of the radial, median, and ulnar nerves.

sEMG and fine wire recordings. Surface EMG was recorded from wrist flexors (FCR and FCU), wrist extensors (ECRb and ECU), and hand muscles (APB and ADM). All muscles were located using manual muscle testing at recommended sites (17, 18). ECRb muscle location was ∼45% of the length of the radius as measured from the styloid process (10, 19). FCR muscle location was ∼8–9 cm from the medial epicondyle (10), FCU (∼5–6 cm from the midpoint between the medial epicondyle and the olecranon), ECU (∼9–10 cm from olecranon), ADM (approximately midpoint between the head of the ulnar and the base of the first phalanx of the little finger), and APB (∼1–2 cm proximal of the radial side of the base of the first phalanx of the thumb). The bipolar Ag/AgCl surface electrodes (16 mm diameter) were attached to the skin over the belly of the muscles with an interelectrode distance of 2 cm (center to center). For fine wire recordings, branched bipolar electrodes comprising stainless steel (50 μm diameter; California Fine Wire, Grover Beach, CA) were used. A 2 mm length of insulation was removed from the tip of each wire, and the distal 3 mm of the wire was bent to create a hook. Single wires were inserted into the barrel of 27-gauge (10 mm) disposable needles and autoclaved for sterilization. Two needles were used to insert the two wires, using sterile procedures, into each forearm muscle (interelectrode distance = 10 mm; FCR, FCU, ECRb, and ECU). The location of the muscles was identified as described for surface EMG. All EMG and force data were sampled at 10 kHz with a National Instrument A/D board and custom-written software. EMG signals were band-pass filtered (10–1,000 Hz) and amplified (gain: 100) with Grass P5 series amplifiers.

Peripheral nerve stimulation. Compound muscle action potentials (CMAP; M-waves) were elicited via electrical nerve stimulation using a bipolar felt pad electrode probe and measured for all muscles at rest. A Digitimer stimulator (model D57A) was used to produce a single electrical stimuli (duration 200 μs) at randomized interstimulus intervals between 3.5 and 4.5 s to the radial, median, and ulnar nerve at recommended sites (17) around the elbow joint with the cathode placement distal to the anode. The stimulation site of the radial nerves was approximately at the spiral groove of the humerus, the median nerve was located approximately medial to the brachial artery and biceps tendon, and the location of the ulnar nerve was approximately posterior to the midpoint between the olecranon and the medial epicondyle of the humerus. The electrode probe was carefully maneuvered to locate the site that resulted in evoked potentials at the lowest possible current. The lowest threshold position was then marked on the skin with a nonpermanent marker and held constant by the same examiner throughout the experiment. An external fixation system was not used because of the difficulty in positioning these electrodes in a manner that could obtain maximal M-waves at currents low enough to avoid cross-nerve stimulation. Stimulation intensity was increased incrementally in steps of 2 mA from 4 to 22 mA and in steps of 5 mA from 25 to 50 mA for each nerve in random order.

Maximal voluntary contraction. The peak torque of three trials measured during a slow ramp to maximal voluntary contraction (MVC) in the horizontal and vertical planes (wrist flexion, wrist flexion, radial deviation, and ulnar deviation) was recorded. The subjects were instructed to increase torque slowly and steadily over 2.5 s and sustain a MVC for an additional 1.5 s. Audio support that consisted of a linearly increasing tone (frequency = 400–737.5 Hz) followed by a continuous tone (frequency: 900 Hz), visual feedback, and verbal encouragement were provided to assist the subject with the task. Force was recorded via 6-df force transducer (JR3 45E15A-163-A400N60S, Woodland, CA).

Data analysis. M-waves, rms EMG values, and force data were sampled with a 16-bit National Instruments A/D board operated by a computer running custom-written Labview program (LabVIEW, Version 8.2.1; National Instruments). All data were analyzed offline using a custom-written Labview program. None of the data contained prestimulus voluntary EMG. The peak to peak amplitude of the responses in all six muscles to radial, median, and ulnar nerve stimulation was obtained at each intensity. Cursors were placed in accordance to the latency of the M-wave of the target muscle to measure the amplitude of cross-talk responses for nontarget muscles (see Fig. 1A). The latency and characteristics of these responses were carefully monitored to exclude any contribution from reflex activity. H-reflex (the earliest possible reflex activity) in FCR was recorded in eight subjects during median nerve stimulation. In addition, three subjects displayed reflex activity in a nontarget muscle (FCU) also during median nerve stimulation. The latency of these reflexes was ∼18 ms. All peak amplitudes included in the measurement of cross-talk responses occurred prior to any of these reflexes. Examples of reflex activity in target and nontarget muscles together with cross-talk responses are revealed in Fig. 1, B and C. The peak to peak amplitude was then normalized to the maximum amplitude (Mmax) produced when the innervating nerve of each muscle was stimulated during the incremental nerve stimulation process that reached an average supramaximal intensity of between 125 and 175% of intensity needed to elicit Mmax (i.e., the "true" Mmax). Rms EMG values for each of the forearm muscles were calculated at MVC during the slow ramp to maximum contraction in all four directions as described above. Cursors were placed 10% below and above the MVC value to calculate rms EMG at MVC.

Statistical analysis. The means and standard error of the CMAP, normalized to Mmax, were calculated for all six muscles in response to stimulation of all three nerves (n = 12). The data from four subjects who showed evidence of cross nerve stimulation (i.e., stimulation of a nontarget nerve; e.g., ulnar nerve stimulation during intended
median nerve stimulation) were not included in the main results. The evidence of such cross nerve stimulation is presented separately in the RESULTS. Pearson product-moment correlation coefficients were calculated to describe the relationship between the increases in M-wave of the target muscles in relation to the size of cross-talk in the nontarget muscles. The amplitude of responses to nerve stimulation obtained in the forearm muscles from fine wire recordings are presented for three subjects. The mean rms EMG activity in each of the four wrist muscles during maximal voluntary contractions in each direction was expressed as a percentage of its own maximum muscle rms activity. A two-way repeated-measures ANOVA (voluntary vs. evoked context/muscle) was used to make comparisons between cross-talk arising from stimulus-evoked responses and that arising from voluntary contraction. Cross-talk signals were defined as those recorded in any of the four muscles during stimulation of a noninnervating nerve (i.e., FCR and FCU during radial nerve stimulation; ECRb, FCU, and ECU during median nerve stimulation; and ECRb, FCR, and ECU during ulnar nerve stimulation) and any voluntary EMG signals recorded from a muscle during MVC in a direction greater than 90° from its preferred direction (i.e., FCR and FCU during extension contraction, ECRb and FCR during ulnar deviation, FCU and ECU during radial deviation, and ECRb and ECU during flexion contraction). Alpha was set at $P < 0.05$.

RESULTS

**sEMG cross-talk in response to stimulus-evoked potentials.** Cross-talk was observed in all nontarget muscles in response to the activation of target muscles produced by radial, median, and ulnar nerve stimulation (for evidence see Fig. 2, A–C). The figures represent a descriptive display of the recruitment curves generated for response amplitude vs. stimulation intensity for all muscles recorded. For example, in Fig. 2A, the response amplitude recorded in radial nerve innervated muscles, ECRb, and ECU represents the stimulus-evoked potentials of the muscles, whereas the response amplitude recorded in nonradial nerve innervated muscles (e.g., FCR and FCU) is cross-talk. From Fig. 2, close similarity in the shape of the recruitment curves for both the target and nontarget muscles is evident. This is consistent with a direct relationship between the sizes of the cross-talk response in the nontarget muscles as a function

![Fig. 1](http://jap.physiology.org/)

Raw traces of individual subjects during median nerve stimulation at specific stimulation intensities. Long dashed lines represent the latency at which cursors were placed to measure M-waves and cross-talk. A: raw traces of all 6 muscles at maximal amplitude (Mmax) of flexor carpi radialis (FCR). B: raw traces displaying evidence and latency of H-reflex in target muscle (FCR) at 2 low stimulation intensities (i.e., 4 and 6 mA). C: raw traces of the presence of reflex activity in nontarget muscle [flexor carpi ulnaris (FCU)] at 40 and 45 mA. [Note, all peaks in the measurement of M-wave and cross-talk occurred prior to H-reflex (the earliest possible reflex contribution) and any other reflex activity.] Arrow, stimulus artifact. Calibration bars: 1 mV and 15 ms. ECRb, extensor carpi radialis brevis; ECU, extensor carpi ulnaris; APB, abductor pollicis brevis; ADM, adductor digit minimi.
of increased activation of the target muscles. A strong correlation was found between the response amplitude of target muscles and their neighboring nontarget muscles for stimulation of all three nerves (all, \( r > 0.96, P < 0.05 \); see Fig. 2).

Table 1 presents the maximum amplitude of EMG responses for all target and nontarget muscles when the target muscle with the lower threshold level was at Mmax (i.e., ECRb for radial, FCR for median, and FCU for ulnar nerve stimulation) in response to stimulation of the three nerves. The size of the responses is presented as a percentage of the true Mmax for each muscle. All nontarget muscles recorded some degree of sEMG cross-talk, with the highest amount recorded in the FCU muscle as a consequence of median nerve stimulation (39.4%) and lower amounts for ECRb and ECU responses to median nerve stimulation. There was also more than 20% of sEMG cross-talk recorded from ECU and APB muscles with ulnar nerve stimulation. EMG signals recorded from fine wire EMG from three subjects also provide evidence of the presence of EMG cross-talk (Table 1). The degree of EMG cross-talk with fine wire EMG was, however, much smaller than sEMG in all instances.

**Stimulus-evoked sEMG cross-talk vs. sEMG produced by voluntary contraction.** Cross-talk signals were generally comparable for sEMG responses to electrical nerve stimulation (Table 1) and sEMG produced by voluntary contraction (Table 2). There was no significant main effect between voluntary and stimulus-evoked recording contexts (\( F_{1,11} = 0.5806 \) and \( P = 0.46 \)). However, there were individual examples of large stimulus-evoked cross-talk signals in some muscles relative to voluntary contractions. For example, the highest sEMG cross-talk value in either stimulus-evoked or voluntary contractions was recorded in FCU during median nerve stimulation (39.4% of Mmax). Also, the size of the evoked response to radial nerve stimulation in the nontarget FCR was 32.5% of Mmax, whereas the ratio between the voluntary rms EMG recorded during extension and flexion MVCs was 6.7% (\( P < 0.05 \)).

**Cross nerve stimulation.** We were able to identify cross nerve stimulation by the occurrence of a sudden increase in the recruitment gradient of sEMG signals, with increasing stimulation intensity to a noninnervating nerve in both hand and forearm muscles that share the same innervating nerve. This was the typical response for unintended nerve stimulation. Amplitude vs. stimulation intensity plots for two subjects are presented vs. the average group data in Fig. 3 to illustrate evidence of cross nerve stimulation. Data from subject 3 showed a typical response of cross nerve stimulation from the median to the ulnar nerve, with a simultaneous increase in response size in ulnar nerve innervated muscles (FCU in the forearm and ADM in the hand) at an intensity well above the threshold for activating median nerve. Subject 4, however, showed an interesting case of cross nerve stimulation, with a sudden increase in response amplitude only for the ADM muscle of the hand but not for the ulnar innervated FCU. This type of effect might be expected if there were substantial differences in threshold levels for activating the axons of the different muscles, which could in principle be caused by differences in the size or location of axons relative to the stimulation. For this subject, however, a lower recruitment threshold for ADM axons could not be the reason for the

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**Fig. 2.** Descriptive presentation of the recruitment curves of 6 muscle groups. Plots are generated from the amplitude of the responses expressed as a percentage of each muscle’s true Mmax (i.e., produced by stimulation of the innervating nerve) vs. the intensity of nerve stimulation. A: radial nerve stimulation; B: median nerve stimulation; C: ulnar nerve stimulation. Standard error bars are displayed on the graph. Pearson product-moment correlation coefficients; \( r \) represents the relationship between the increases in M-wave of the target muscles and the size of cross-talk in the nontarget muscles. A: FCR and ECRb, FCU and ECU; B: FCR and FCU, APB and ADM; C: FCU and ECU, ADM and APB.
different behavior of the two muscles, as both muscles had low recruitment threshold when the ulnar nerve was stimulated. Another possibility would be the existence of communicating branches between the median and ulnar nerves in that particular subject. This condition is known as Martin-Gruber anastomosis in the forearm and Riche-Cannieu anastomosis in the hand (6). The existence of any one of the many forms of these communicating branch phenomena could account for a sudden increase in sub-pools of muscles innervated by a single nerve.

Further increases in median nerve stimulation intensity beyond levels supramaximal for activating median-innervated muscles could induce cross nerve stimulation for most subjects (i.e., shown by simultaneous increases in response size in FCU and ADM). Figure 4 depicts an example from one subject when the stimulus intensity was increased to the maximal stimulation output with median nerve stimulation. The size of the maximal M-wave recorded in FCU during simultaneous median and (unintended) ulnar nerve stimulation was close to the sum of the response due to cross-talk from median nerve stimulation (i.e., ≈40%) and the Mmax produced by isolated ulnar nerve stimulation (i.e., 100%). Therefore, the sum of ~140% in M-wave size could be recorded as depicted in Fig. 4. Cross nerve stimulation, however, only occurred for median nerve stimulation as we did not find any occurrence of cross nerve stimulation with radial or ulnar nerve stimulation even at high intensity.

Table 1. Average values of sEMG amplitudes recorded from 6 muscles at the lowest Mmax intensity of the innervated muscle in response to electrical nerve stimulation (radial, median and ulnar nerve)

<table>
<thead>
<tr>
<th>Muscles</th>
<th>Nerve Stimulation, % Mmax</th>
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<tbody>
<tr>
<td></td>
<td>Radial Nerve</td>
</tr>
<tr>
<td></td>
<td>Mean sEMG</td>
</tr>
<tr>
<td>FCR</td>
<td>32.5 ± 6.0</td>
</tr>
<tr>
<td>FCU</td>
<td>25.9 ± 3.5</td>
</tr>
<tr>
<td>ECRb</td>
<td>100</td>
</tr>
<tr>
<td>ECU</td>
<td>98.4 ± 0.6</td>
</tr>
<tr>
<td>ADM</td>
<td>1.5 ± 0.2</td>
</tr>
<tr>
<td>APB</td>
<td>3.0 ± 0.8</td>
</tr>
</tbody>
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Values are means ± SE. The results are presented as a percentage of the true Mmax produced by stimulation of the innervating nerve of each individual muscle. Fine wire EMG amplitudes were recorded from 4 forearm muscle groups in 3 subjects and presented as a percentage of the true Mmax. sEMG, surface electromyography; FCR, flexor carpi radialis; FCU, flexor carpi ulnaris; ECRb, extensor carpi radialis brevis; ECU, extensor carpi ulnaris; APB, abductor pollicis brevis; ADM, adductor digitii minimi.

DISCUSSION

We demonstrated that our simple method of stimulating multiple nerves can clearly differentiate the evoked potentials produced by target muscles from signals arising from sEMG cross-talk. The method also provided quantitative information about the potential for cross-talk between muscles of the forearm and hand. The size of the cross-talk recorded in forearm and hand muscles was often large and could lead to incorrect interpretation of evoked responses (e.g., Ref. 16). As a comparison against lower limb muscle cross-talk, the ratio of signals detected on tibial bone area, peroneus brevis, and soleus muscles to those detected on tibialis anterior (TA) muscle during TA motor point stimulation were 19.4, 7.0, and 5.0%, respectively (3). We also showed that the problem of cross-talk arising from stimulus-evoked responses is at least as great as that arising from voluntary contraction. The method can also reveal the occurrence of cross nerve stimulation and could provide a useful methodological check for reflex studies involving peripheral nerve stimulation. Finally, the fact that the amplitude of responses to simultaneous stimulation of multiple nerves (i.e., by inadvertent cross-nerve stimulation) was larger than the true maximal M-wave shows that cross-talk signals can summate with genuine activity from a target muscle. This illustrates the serious nature of the problem presented by EMG cross-talk for interpreting TMS and reflex responses, where contributions from synergist and even antagonist muscles might contribute to the evoked-potentials recorded from electrodes over the target muscle.

In considering the issue of EMG cross-talk via nerve stimulation methods, it is important to confirm that the EMG signals recorded in nontarget muscles are not due to cross nerve stimulation (i.e., inadvertent stimulation of the nerve that innervates a nontarget muscle). This was done by recording and examining the characteristics of two hand muscles that are innervated by the same nerve as the forearm muscles. For example, we recorded minimal EMG signals from both ulnar and median nerve innervated hand muscles (ADM and APB) during radial nerve stimulation, while at the same time there was an increase in EMG signals in the forearm muscles (FCU and FCR) that share the same innervating nerves as the hand muscles. In addition, we observed that EMG signals of both FCU and FCR first appeared at the same intensity as when an
M-wave of the stimulated muscles (ECRb and ECU) appeared. The EMG signals of these nontarget muscles continued to increase in line with the EMG in the target muscles and remained stable in amplitude once the M-wave of the target muscles reached its maximum (see Fig. 2). Nonetheless, we did record cross nerve stimulation in four subjects and at high stimulation intensities; however, this only occurred for median nerve stimulation that inadvertently recruited the ulnar nerve. In addition to cross nerve stimulation, any potential contribution from reflex activity was also considered. To account for this, the latency and characteristics of any potential reflex activity present throughout the experiment was examined (see Fig. 1). A typical example of muscle cross-talk (group data; n = 12) and cross nerve stimulation (subjects 3 and 4) recorded at nontarget ulnar nerve innervated muscles (FCU and ADM) during median nerve stimulation. Group data: FCU, solid line with □; ADM, solid line with □; subject 3: FCU, solid line with ●; ADM, solid line with ▪; subject 4: FCU, solid line with □; subject 4: ADM, solid line with ▪. Y-axis: percentage of the muscle’s true Mmax (i.e., produced by stimulation of the innervating nerve); X-axis: intensity of nerve stimulation. Standard error bars are displayed on the graph for group data.

Comparison with voluntary contraction and fine wire EMG. To establish the importance of identifying sEMG cross-talk arising from noninvasive stimulation techniques, we compared the size of this muscle cross-talk with that arising from voluntary contraction as it has been the focus of many recent studies (2, 7, 9, 11, 23, 24). We recorded from the same subjects, and cross-talk signals from H-reflex is also seen in Fig. 1B). Only peaks in sEMG signals that occurred prior to the earliest possible reflex contribution (i.e., H-reflex) were included into the calculation of cross-talk. We also found that the latency of the reflex recorded in nontarget muscles was approximately similar to H-reflex in target muscles and its size smaller compared with the cross-talk signal size. There was also a strong correlation between the response amplitude of the target muscles and their neighboring nontarget muscles in all three nerve stimulations (see Fig. 2). Thus our measures of sEMG cross-talk were influenced by the size of the responses in the target muscles rather than any reflex activity.

Potential sEMG cross-talk in response to TMS. Our results revealed that the EMG signal recorded in nontarget muscles affected by cross nerve stimulation is larger than the “true” Mmax produced by supramaximal stimulation of the innervating nerve. The size of the EMG signals recorded during simultaneous (inadvertent) stimulation of multiple nerves was close to the sum of sEMG muscle cross-talk signal size and the true Mmax (see Fig. 4). This highlights the seriousness of the problems for interpretation of TMS and reflex studies posed by EMG cross-talk; whereas electrical stimulation can selectively stimulate specific muscle groups, it is difficult to know precisely which muscles might be activated by reflex and TMS methods. Since many reflex and TMS responses are likely to contain contributions from many muscles within functionally related muscle groups, the EMG signals recorded from electrodes over a specific muscle may represent an overestimation of the true response in a target muscle. This may not be a critical error if the evoked responses are interpreted as the product of a muscle group (rather than an individual muscle). However, if more detailed information about the activation of multiple muscles is sought (e.g., agonist vs. antagonist activation), the current results indicate that it is essential to rule out the possible contribution of EMG cross-talk before the data can be validly interpreted.

By using the hand muscles as a control we also found that there was considerable (i.e., 24%) muscle cross-talk recorded at the APB muscle (i.e., a nontarget muscle) when the ulnar nerve was stimulated. This cross-talk between hand muscles was not due to inadvertent stimulation of the median nerve as there was minimal activity in the FCR muscle that shares the same innervating nerve with the APB muscle. In addition, the size of the EMG signal did not continue to increase once the M-wave of the ulnar nerve innervated muscle (i.e., ADM) reached maximum. This evidence of cross-talk in the hand is particularly important as many TMS studies focus on hand muscles due to their strong representation in the primary motor cortex (M1). Therefore, researchers need to consider the problem of cross-talk when conducting TMS studies, and great care is needed when interpreting the sizes of motor evoked potentials (MEPs) in both the hand and forearm.

Fig. 4. Example of cross nerve stimulation for a single subject recorded on nontarget (ulnar nerve innervated muscles, FCU and ADM) during median nerve stimulation at high intensity. FCU, solid line with ■; ADM, solid line with □. Y-axis: percentage of the muscle’s true Mmax (i.e., produced by stimulation of the innervating nerve); X-axis: intensity of nerve stimulation.
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during the same experimental session, and demonstrated that EMG activity was recorded from all muscles that were nonprime movers during voluntary contractions in four directions (Table 2). The amount of EMG activity recorded from these muscles is likely to be due to a combination of sEMG cross-talk and of genuine activity through a role as an antagonist, synergist, or stabilizer. Although the voluntary activity recorded from nonprime movers could arise due to the sum of each of these factors, there was a comparable degree of sEMG activity in each muscle relative to the sEMG muscle cross-talk arising from electrical nerve stimulation. Thus the potential for cross-talk arising from electrical stimulation is at least as great as that arising from voluntary contraction, at least for the muscles of the forearm. Therefore, it is particularly important to account for sEMG muscle cross-talk produced by noninvasive stimulation techniques, as it may pose more serious problems for misinterpretation of data compared with voluntary contraction.

It was previously proposed that the use of fine wire EMG is necessary to identify the presence of muscle cross-talk in a wide range of voluntary muscle contractions (2). However, the utility of intramuscular techniques in identifying muscle cross-talk during noninvasive stimulation studies is less obvious. In our study, we used fine wire EMG on three subjects to test if the technique provided any additional information in identifying the presence of sEMG cross-talk and if cross-talk could be completely removed when using fine wire recordings. We found that cross-talk was still present between muscles of the forearm with fine wire EMG. The nature of muscle cross-talk observed was of a similar pattern to that of surface EMG, but was much smaller in all muscle groups. Nevertheless, the presence of muscle cross-talk must still be accounted for when using fine wire EMG. Therefore in light of this issue, the technical difficulties in using fine wire EMG, and the lack of global information acquired, we suggest that the present method using sEMG is an appropriate method for identifying and quantifying the potential for cross-talk in both stimulus-evoked and voluntary contraction contexts.

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AUTHOR CONTRIBUTIONS
Author contributions: V.S.S., S.R., and T.J.C. conception and design of research; V.S.S. performed experiments; V.S.S. and T.J.C. analyzed data; V.S.S., S.R., and T.J.C. interpreted results of experiments; V.S.S. prepared figures; V.S.S. drafted manuscript; V.S.S., S.R., and T.J.C. edited and revised manuscript; V.S.S., S.R., and T.J.C. approved final version of manuscript.

REFERENCES

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