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Motor adaptation varies between individuals in the transition to sustained pain

Simon J Summers[1], Lucy S Chipchase[1,2], Rogerio Hirata[3],

Thomas Graven-Nielsen[4], Rocco Cavaleri[1], Siobhan M Schabrun[5]

1. School of Science and Health, Western Sydney University, NSW 2560, Australia
2. Faculty of Health, University of Canberra, ACT, Australia
3. Department of Health Science and Technology, Aalborg University, Aalborg, Denmark
4. Center for Neuroplasticity and Pain (CNAP), SMI, Department of Health Science and Technology, Faculty of Medicine, Aalborg University, Aalborg, Denmark.
5. Neuroscience Research Australia, NSW, Australia

Corresponding author:

Dr. Siobhan Schabrun

Neuroscience Research Australia

Margarete Ainsworth Building, 139 Barker street (PO box 1165)

Randwick, NSW 2031, Australia

Phone: +61 421568494

Email: s.schabrun@neura.edu.au

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ABSTRACT

Musculoskeletal pain is associated with altered motor control that despite short-term benefit, is hypothesised to have long-term consequences, contributing to the development of chronic pain. However, data on how motor control is altered when pain is sustained beyond a transient event are scarce. Here, we investigated motor adaptation, and its relationship to corticomotor excitability, in the transition to sustained muscle pain. Twenty-eight healthy individuals were injected with nerve growth factor (NGF) into the right extensor carpi radialis brevis (ECRB) muscle on Days 0 and 2. Motor adaptation and corticomotor excitability were assessed on Day -2, prior to injection on Days 0 and 2, and again on Days 4 and 14. Motor adaptation was quantified during a radial-ulnar movement as kinematic variability of wrist flexion-extension and pronation-supination, and as electromyographic (EMG) variability of ECRB activity. Pain, muscle soreness, and functional limitation were assessed from Days 0-14. Pain, muscle soreness and functional limitation were evident at Days 2 and 4 ($p<0.001$). EMG variability reduced at Days 4 and 14 ($p<0.04$), with no change in kinematic variability ($p=0.9$). However, data revealed variation in EMG and kinematic variability between individuals: some displayed increased motor variability while others a decrease. Individuals who displayed an increase in EMG variability following four days of pain also displayed an increase in corticomotor excitability ($r=0.43$, $p=0.034$). These findings suggest individual adaptation of the motor system in the transition to sustained pain that could have implications for clinical musculoskeletal pain disorders.

Keywords: Motor cortex plasticity, Musculoskeletal pain, Transcranial magnetic stimulation, Motor variability, Motor adaptation

INTRODUCTION

Musculoskeletal pain is associated with adaptations in motor control [2,34,60]. In the short-term, altered motor control (or motor adaptation) is considered a beneficial response serving to protect the body from further pain or injury [35]. However, in the long-term, altered motor control is hypothesised to contribute to the maintenance or recurrence of pain [35,47,61]. Although numerous cross-sectional studies have documented motor adaptation in acute and chronic pain [32,36,44,91,94], few studies have characterised motor adaptation in the transition to sustained pain. Understanding motor adaptation, and the underlying mechanisms, as pain develops may have implications for understanding why some people fail to recover from musculoskeletal pain.

A spectrum of motor control changes have been documented in people with acute and chronic pain ranging from subtle changes in muscle activity to complete movement avoidance [35,34]. Although there is evidence of individual-specific motor adaptation [33,78,87], the most consistent finding in response to acute experimental pain (lasting 7-15 minutes) is an increase in motor variability (i.e. greater variation in muscle activation, motion of joints and force applied during an isometric contraction [32,45,48,79]). However, in studies of people with chronic neck, shoulder, back, and knee pain, decreased motor variability has been observed [22,28,31,44,48,55]. One interpretation is that an increase in motor variability during acute pain reflects the search for a motor strategy that maximises task performance while minimising pain, while in chronic pain, reduced variability reflects an avoidance strategy that limits painful movements and postures [6,57,79]. However, in the long-term, reduced variation in motor output is thought to increase accumulative load on tissues, increasing the risk of further pain and/or injury [35,79]. Opposing changes in motor

variability in the acute and chronic stages of pain suggest motor adaptation varies as a function of pain duration.

Only one study has examined motor adaptation as pain progressively develops and is sustained over time [53]. Compared to controls, force direction, but not force variation, was greater during an isometric wrist-extension contraction when pain lasted up to four days. However, variability in muscle activation and movement kinematics were not explored. Further, no study has investigated the relationship between motor adaptation and corticomotor excitability in the transition to sustained pain despite evidence for a relationship in cross-sectional studies. For example, in chronic back pain, a posterior and lateral shift and an increase in excitability of the corticomotor representation of the transverse abdominis muscle was associated with a delay in activation of this muscle during arm movement [85].

Here, we used repeated intramuscular injection of nerve growth factor to induce muscle pain that progressively developed over 4 days in order to investigate i) the nature and time-course of motor adaptation (variability in muscle activation and movement kinematics) and ii) the relationship between motor adaptation and corticomotor excitability, in the transition to sustained muscle pain. Based on previous studies [72,79], it was hypothesised that motor variability would increase when pain was sustained for four days, and that the increase in motor variability would be associated with increased corticomotor excitability.

METHODS

Participants

Twenty-eight healthy individuals (mean ± standard deviation [SD] age 23 ± 4 years; 13 females) participated. All participants were right handed, verified by the Edinburgh Handedness Inventory [62]. Participants had no history of neurological or upper limb conditions and completed a transcranial magnetic stimulation (TMS) safety screen prior to study commencement [39]. All participants provided written, informed consent in accordance with the Declaration of Helsinki. The institutional Human Research Ethics Committee approved the study (H11949). Secondary outcomes from this protocol are published elsewhere and include the effects of NGF-induced pain on joint position sense [80].

An *a priori* sample size calculation was performed for the primary aim using GLIMMPSE software [43] with data from a previous NGF and transcranial magnetic stimulation (TMS) study [72]. These data demonstrate a mean (SD) change in map volume of 2 (1.2) mV at Day 4 following two NGF injections. Using these values, 25 subjects were needed to identify a minimum change in map volume of 2 (1.2) mV at Day 4 with a power of 80% at the 0.05 significance level.

Experimental protocol

Each participant attended the laboratory on five occasions: Day -2, 0, 2, 4, and 14 (Figure 1). As this study was focussed on motor adaption as pain developed and peaked (Days 0-4), rather than recovered (Days 5-14), test intervals providing higher temporal resolution in the early stage of the model were selected. Indeed, studies have shown that pain progressively develops and peaks at Day 4 following 2 injections of NGF before gradually resolving [19,30,72]. In addition, studies have shown that injection of NGF does not produce

acute symptoms, with pain and soreness first beginning to develop 3-hours post-injection [1,30,59,82]. For this reason, we did not include an outcome assessment timepoint immediately following NGF injection.

On Days 0 and 2, nerve growth factor (NGF) was injected into the muscle belly of the right extensor carpi radialis brevis (ECRB) muscle. At the beginning of each session, maps of the corticomotor representation of the right ECRB muscle were obtained using transcranial magnetic stimulation (TMS). Pain intensity (numerical rating scale, NRS), muscle soreness (Likert scale), and functional limitation (patient rated tennis elbow evaluation, PRTEE) were recorded on Days 0, 2, 4, and 14. To examine motor variability, wrist angle kinematics and electromyographic data of the ECRB muscle during a wrist radial/ulnar deviation task were measured at each test session (Days -2, 0, 2, 4, and 14). The Day -2 (two days before Day 0) test session was included to assess the test-retest reliability of performance on the radial/ulnar deviation task and corticomotor outcomes in the absence of pain (Day -2 versus Day 0).

NGF-induced muscle pain

Sterile recombinant human NGF (5 µg, 0.2 ml) was injected into the muscle belly of the right ECRB muscle using a 1-mL syringe with a disposable needle (27G) [30,72]. The site of injection was determined by identifying the position 1 cm lateral to a point 5 cm distal to the lateral epicondyle [7]. Palpation of the muscle belly of ECRB during resisted wrist extension and radial deviation confirmed the injection site. A mark was drawn around the injection site with a permanent marker to ensure consistent placement of the injection in the ECRB muscle across sessions. Injection of NGF at this site has been shown to induce sustained lateral elbow pain [7,19,72], with the experience of pain and soreness similar to that reported by patients with lateral epicondylalgia of approximately 26 weeks duration [7].

Assessment of pain, muscle soreness, and functional limitation

An 11-point numerical rating scale (NRS) (0 = no pain; 10 = worst pain imaginable) was used to assess pain intensity. Muscle soreness was assessed using a modified 7-point Likert scale: "0 = complete absence of soreness; 1 = light soreness in the muscle felt only when touched/vague ache; 2 = moderate soreness felt only when touched/a slight persistent ache; 3 = light muscle soreness when lifting or carrying objects; 4 = light muscle soreness, stiffness or weakness when moving the wrist without gripping an object; 5 = moderate muscle soreness, stiffness or weakness when moving the wrist; and 6 = severe muscle soreness, stiffness or weakness that limits the ability to move" [30,77].

The Patient-rated tennis elbow evaluation (PRTEE) was used to assess average pain and functional limitation. Scores for pain (5 items) and function (10 items) were combined to provide a total score ranging from 0 (no pain and no functional limitation) to 100 (worst pain imaginable with significant functional limitation) [46]. The PRTEE has excellent test-retest reliability (r = 0.77 – 0.93) and correlates well with the Disabilities of Arm, Shoulder, and Hand (DASH) Questionnaire in a persistent elbow pain population (r = 0.75) [69].

Experimental set-up for assessment of motor variability

An adapted version of the wrist radial/ulnar deviation task developed by Bergin et al. [6] was used to measure motor variability. This task was selected for two reasons. First, it was anticipated that changes in the excitability of corticomotor projections to ECRB, as a result of NGF-induced pain, would relate to movements at the wrist that involve the primary action of ECRB (i.e. radial deviation) [6]. Second, this task has been demonstrated to measure motor variability at the wrist during acute forearm extensor pain [6] and thus

provides an established model to study the relationship between sustained forearm extensor pain, motor variability, and corticomotor excitability.

To assess the kinematics of the right (injected) arm during the radial/ulnar deviation task, a two-segment model was used to 3D model the right forearm and hand [68]. Six active markers were positioned over the segments: 1) three reflective markers attached to the dorsum of the right hand between the head of the second and fifth metacarpal bones, and 2) three reflective markers attached to the dorsum of the forearm 2 cm proximal to the radial styloid (Figure 2A). Six virtual markers were digitised to identify the medial and lateral epicondyle, ulna and radial styloid, and head of the second and fifth metacarpal bones. These virtual markers were used to create the 3D model of the forearm (medial and lateral epicondyle, ulna and radial styloid) and hand (ulna and radial styloid, head of the second and fifth metacarpal bones). Based on the 3D model, joint angles for wrist radial-ulnar deviation, forearm pronation-supination, and wrist flexion-extension were calculated [26]. Movements of the clusters were recorded at 200 Hz by one position sensor containing three cameras (Optotrak Certus System, Northern Digital Inc., Waterloo, Canada) and processed using Visual 3D (C-Motion, Version 4, Germantown, MD).

To assess muscle activation of ECRB during the radial/ulnar deviation task, surface EMG recordings were collected using a bipolar silver/silver chloride surface electrode positioned over the muscle belly. Prior to application, the skin was lightly abraded using Nuprep skin prep gel (Weaver and Company, Colorado, USA), then cleaned with alcohol. The EMG signals were band-pass filtered (20–450 Hz) and sampled at 1000 Hz (Zerowire EMG system, Aurion, Zerowire, Italy).

Participants were seated in an upright position with the right forearm resting on a horizontal platform in mid-position between pronation and supination with the elbow in approximately 90° flexion. The forearm was stabilized using an adjustable brace at the forearm and an adjustable clamp proximal to the wrist (Figure 2A). This ensured that participants started in the same position across experimental sessions, allowed unconstrained wrist motion and forearm rotation, and prevented upper limb movements from affecting performance during the radial/ulnar deviation task. The apparatus (Figure 2A) was fixed to a table, and the table was secured with a chair to the floor to ensure seat position was controlled over each experimental session.

In the first experimental session (Day -2), the neutral position of the wrist, and maximal range of motion for radial and ulnar deviation were recorded. The neutral position was measured using a handheld goniometer, while maximal radial and ulnar range of motion were recorded using a laser pointer and a paper board. A small foam block with two laser pointers were attached to the hand of each participant (Figure 2A). Once the neutral position of the wrist was set, a blank paper board was placed 60 cm in front of the hand and the position of the active laser on the paper board was marked corresponding to the 'neutral position'. To identify maximal radial and ulnar range of motion, the position of the active laser on the paper board was marked in maximal radial and ulnar deviation. In addition to the active laser, the position of the reference laser was marked and used to ensure consistent orientation of the hand in the neutral position before commencing the task (Figure 2B). After the hand was orientated in wrist neutral, the reference laser was switched off during the experimental procedure. The paper board set for each participant was used across sessions to ensure reliable assessment of the experimental trials.

Procedure for assessment of motor variability

The experimental task involved repeated radial/ulnar deviation of the wrist between two target angle regions that were marked on the paper board (Figure 2B). Participants were instructed to move the active laser as accurately as possible between two target regions: 80-100% of their maximal radial deviation range to 20-40% of their maximal ulnar deviation range in time with a metronome (90 beats per minute) [6]. We used a metronome-based task as motor variability has been shown to change as a function of movement speed [3,9,18,21], thus ensuring any changes in motor variability could be attributed to pain and not to participants adjusting their movement speed across sessions. The targets were standardized to a percentage of maximal range to account for individual differences in maximal radial/ulnar deviation range of motion.

At the start of each session, participants practised until they completed the task at the correct frequency. Data from the practice trial were not recorded. Emphasis was placed on moving to each target as accurately as possible. Forty-five repetitions were recorded starting and finishing in the neutral position before (Day -2 and Day 0 sessions), during (Day 2 and Day 4 sessions), and after a period of sustained elbow pain (Day 14 session). Participants were asked at the end of each 45-repetition trial whether they perceived fatigue in their forearm or wrist.

Corticomotor excitability

Electromyography (EMG) was recorded from the right ECRB (electrode type and placement as described above). The ground electrode was placed over the right olecranon. The EMG signal was pre-amplified 1000 times, band pass filtered between 20 and 1000 Hz, and sampled at 2 kHz using Signal 3 software and Power 1401 data acquisition system (Cambridge Electronic Design, Cambridge, UK).

Single-pulse, biphasic stimuli were delivered using a Magstim Super Rapid^2 (Magstim Co. Ltd, Dyfed, UK) and a 7-cm figure of eight coil. The coil was positioned tangentially to the scalp with the handle pointing posteriorly at 45° from midline. This orientation is optimal for the induction of posterior-to-anterior (PA) directed current and activation of horizontal cortical connections in M1 [5,13]. The optimal scalp site ('hotspot') for evoking responses in the ECRB muscle was then established by systematically moving the coil in 1 cm increments around the motor cortex. The site that evoked the largest EMG responses at a given stimulator intensity was considered the hotspot. The stimulus intensity for mapping was set at 120% of the resting motor threshold (rMT), defined as the minimum stimulator intensity at which 5 out of 10 stimuli applied at the hotspot evoked a response with a peak-to-peak amplitude of at least 50 μV [27]. This intensity was determined on Day -2 and kept constant on Days 0, 2, 4 and 14.

Participants were fitted with a tight silicon cap marked with a 1 x 1 cm grid positioned and orientated to the vertex. The vertex was identified using the International 10/20 system [42]. Five TMS pulses were applied, with an inter-stimulus interval of five seconds, at each site of the grid [15,93]. The number of scalp sites stimulated was pseudorandomly increased until no MEP was evoked in all border sites [73]. A

neuronavigation system (Brainsight2, Rogue Resolutions Ltd, Cardiff, UK) was used in conjunction with the silicon cap to ensure accurate coil placement at each grid site. Participants were instructed to maintain their right hand and forearm relaxed with their wrist pronated during the experiment. Trials that presented with background EMG activity were discarded (<3% of trials). All TMS procedures adhered to the TMS checklist for methodological quality [17].

Map volume, single site excitability, and centre of gravity (CoG) were calculated. Map volume was calculated as the sum of all active sites. A site was considered 'active' when the mean peak-to-peak MEP amplitude of the five MEPs evoked at that site was greater than 50 μV. Single site excitability was calculated as the mean peak-to-peak MEP amplitude of the five MEPs delivered at the 'hotspot'. The centre of gravity was defined as the amplitude-weighted center of the map [90,92], and was calculated using the following formula: CoG = $\sum Vi \; x \; Xi / \sum Vi$, $\sum Vi \; x \; Yi / \sum Vi$; where: Vi = mean MEP amplitude at each site with the coordinates Xi , Yi. Test-retest reliability and validity of these procedures for calculating volume and centre of gravity for upper limb muscles has been previously demonstrated [49,58,90]. Specifically, studies have shown between session reliability for map volume and centre of gravity measures with a 24 hour, 4-day, 1-week, or 2-week inter-session interval [49,51,58,90].

Finally, the number of scalp sites over which TMS evoked a 'discrete peak' in activity in the corticomotor representation was determined. Using an established method, discrete peaks were identified if the average MEP amplitude at a grid site was greater than 50% of the maximum MEP amplitude, separated by MEP amplitudes that were at least 5% lower than

peak MEP amplitudes in 7 out of 8 surrounding grid sites, and were separated by at least 1 grid site from another discrete peak [50,72,74,75].

Data analysis

To determine whether subjects accurately moved the laser pointer to each target zone, video data of the laser was recorded during each trial and analyzed offline using MATLAB R2016b (Mathworks, Natick, MA, USA). Movement accuracy was expressed as a percentage and represented the proportion of repetitions within each experimental session in which participants successfully moved the laser to the radial and ulnar deviation target zones (see figure 3 for representative data). To analyse kinematic and EMG variability, events were created during the analyses of each 45-reptition trial to signify movement cycles – a movement cycle was defined as the period between the starting angle of radial/ulnar deviation (wrist neutral) and when the hand passed through wrist neutral following movements to both radial and ulnar deviation targets). Kinematic variability was quantified as the mean standard deviation in both wrist flexion-extension and forearm pronation-supination across each movement cycle in the 45-repetition trial.

The EMG signals of ECRB were rectified, filtered (low-pass filtered at 25 Hz using a second-order Butterworth filter) and normalized to the peak EMG signal recorded for ECRB during a maximum voluntary contraction of handgrip (performed with participants seated upright with the elbow in approximately 90° degrees) in each experimental session. The mean standard deviation of the normalized root mean squared (RMS) values of ECRB across each movement cycle (defined above) in the 45-repetition trial were then used as a measure of the amplitude variability in surface EMG activation during the task. Thus, the degree of

kinematic and EMG variability was assessed at a whole-cycle level, but not with respect to sub-events within the cycle [29].

Statistical analysis

Statistical Package for the Social Sciences (SPSS) software (version 23 IBM Corp, Armonk, NY, USA) was used for statistical analysis. All data were assessed for normality using visual inspection (Q-Q plot) and Shapiro-Wilk's test. Paired sample t-tests were performed to compare learning effects and stability of corticomotor outcomes over two days in the absence of pain (Day -2 and Day 0). If normality was violated for the paired samples t-test, a Wilcoxon signed-rank test was used. Intraclass correlation coefficients (ICC, two-way mixed effects) were performed to assess test re-test reliability of corticomotor (volume, discrete peaks, CoG) and motor adaptation (EMG and kinematic variability) outcomes over the two baseline experimental sessions (Day -2 and Day 0). ICC values were interpreted as: poor (<0.50), moderate (0.50-0.65), good (0.65-0.80) or excellent (>0.80) [14,65]. Corticomotor outcomes (map volume, single site excitability, discrete peaks, CoG), movement accuracy, and EMG and kinematic variability data were compared between Days 0, 2, 4, and 14 using a one-way repeated-measures analysis of variance (ANOVA). Data that did not meet assumptions of normality were log-transformed, and the Greenhouse-Geisser Correction was applied if data did not meet the assumption of sphericity. Where appropriate, post-hoc analyses were performed using Holm-Sidak multiple comparison tests. Pain (NRS scores), muscle soreness (Likert scale), and functional limitation (PRTEE scores) were compared between Days 0, 2, 4, and 14 using the Friedman test, and if significant, post-hoc comparisons were performed using the Wilcoxon signed-rank test. Pearson correlations (or spearman correlations if data did not meet assumptions of normality) were used to assess the relationship between corticomotor outcomes (volume, discrete peaks, CoG) and EMG and

kinematic variability in the presence of pain (Day 2 and Day 4) and after pain subsided (Day 14). Statistical significance was set at $P< 0.05$.

RESULTS

Assessment of reliability of task performance and corticomotor outcomes in the absence of pain

Motor variability and performance accuracy did not differ (EMG variability: $t_{24} = -1.5$, $p = 0.2$; kinematic variability: $t_{24} = -0.6$, $p = 0.5$; performance accuracy: $Z = -0.72$, $p = 0.5$) between the two pre-pain baseline sessions (Day -2 vs. Day 0). Similarly, corticomotor outcomes were stable when compared between Days -2 and 0, with discrete map peaks ($Z = 0.1$, $p = 0.9$), volume ($Z = 1.1$, $p = 0.3$), and CoG (Latitude: $Z = -1.7$, $p = 0.7$; Longitude: $Z = 1.1$, $p = 0.3$) not significantly changed over time (Table 1). Good to excellent test-retest reliability was demonstrated for all corticomotor (CoGx: ICC = 0.93, $p<0.001$; CoGy: ICC = 0.67, $p<0.001$; MapVol: ICC = 0.86; $p<0.001$) and motor variability (Kinematic: ICC = 0.75, $p<0.001$; EMG: ICC = 0.86, $p<0.001$) outcomes between Days -2 and 0, with the exception of the discrete peak variable where test-retest reliability was poor (ICC = 0.46, $p = 0.1$). Consequently, the discrete peak variable was removed from further analysis.

Pain, muscle soreness, and functional limitation

Three individuals did not develop pain in response to NGF injection and were excluded from analyses. Repeated injection of NGF resulted in pain (Friedman: X^2 (3) = 67.1, $p<0.001$; Figure 4A), muscle soreness (Friedman: X^2 (3) = 66.5, $p<0.001$; Figure 4B), and functional limitation (Friedman: X^2 (3) = 66.6, $p<0.001$; Figure 4C). Post-hoc analysis demonstrated pain and muscle soreness that was present at Day 2 (pain: day 0 vs. 2: $p<0.001$; soreness: day 0 vs. 2: $p<0.001$), remained elevated at Day 4 (pain: day 0 vs. 4: $p<0.001$;

soreness: day 0 vs. 4: p<0.001) and returned toward baseline at Day 14 (pain: day 0 vs. 14: p = 0.1; soreness: day 0 vs. 14: p = 0.1). Similarly, functional limitation was present at Day 2 (day 0 vs. 2: p<0.001), persisted at Day 4 (day 0 vs. 4: p<0.001), and returned to baseline at Day 14 (day 0 vs. 14: p = 0.1). No participant reported fatigue during the experimental protocol.

Influence of sustained muscle pain on motor variability

Attainment of the goal in the radial and ulna deviation direction was not affected by sustained muscle pain, with the proportion of successful repetitions unchanged over time (ANOVA: $F_{2.1, 51.5} = 0.8$, p = 0.5; Day 0 = 99%, Day 2 = 99%, Day 4 = 96%, Day 14 = 99%). At the group level, EMG variability (ANOVA: $F_{2.2, 54.4} = 7.1$, p = 0.001; Figure 5A), but not kinematic variability (ANOVA: $F_{2.3, 62} = 0.1$, p = 0.9; Figure 5B), was altered in response to sustained muscle pain. EMG variability was reduced at Day 4 (post-hoc: day 0 vs. 4: p = 0.04) and Day 14 (post-hoc: day 0 vs. 14: p = 0.01) relative to baseline, but was unchanged at Day 2 (post-hoc: day 0 vs. 2: p = 0.6). However, visual inspection of the data revealed clear variation in both EMG and kinematic variability between individuals, with some individuals displaying increased motor variability at Days 2 and 4 (Day 2, EMG: n=11; kinematic: n=11; Day 4, EMG: n=9; kinematic: n=13; Figure 5A and 5B;) while remaining subjects showed a decrease in variability on both days. Of the 11 individuals that displayed increased EMG and kinematic variability at Day 2, nine (82%) and seven (64%) also displayed an increase in variability at Day 4, respectively. Of the 14 individuals that displayed decreased EMG and kinematic variability at Day 2, nine (82%) and 12 (86%) also displayed a decrease in variability at Day 4 respectively, suggesting a high level of consistency in the motor strategy adopted by an individual over time. Further, across Days 2, 4, and 14, individuals that displayed increased EMG variability also displayed increased kinematic variability (and vice

versa) (r = 0.55, p = 0.004). Post-hoc comparison, using Pearson correlations, of pain (NRS scores), muscle soreness (Likert scale), and functional limitation (PRTEE scores) between those who displayed increased vs. decreased EMG and kinematic variability at Days 2 and 4 did not reveal any associations (all: p>0.12).

Influence of sustained pain on corticomotor excitability and relationship to motor variability

Relative to baseline, map volume was reduced at Day 2 (ANOVA: $F_{2.1, 51} = 4.1$, p = 0.02; post-hoc: day 0 vs. 2: p = 0.02) and Day 4 (post-hoc: day 0 vs. 4: p = 0.003), and returned to baseline at Day 14 (post-hoc: day 0 vs. 14: p = 0.9; Figure 5C). Corticomotor excitability at the hotspot followed the same pattern (ANOVA: $F_{2.2, 51} = 4.2$, p = 0.01) with a reduction in excitability at Day 2 (post-hoc: day 0 vs. 2: p = 0.04) and Day 4 (post-hoc: day 0 vs. 4: p = 0.004), returning to baseline at Day 14 (post-hoc: day 0 vs. 14: p = 0.7). There was no change in the map CoG over time (Latitude: ANOVA: $F_{3, 72} = 0.5$, p = 0.7; Longitude: ANOVA: $F_{3, 72} = 0.6$, p = 0.6). Individuals who displayed a reduction in map volume following four days of sustained pain also displayed a reduction in EMG variability, whereas individuals who increased map volume displayed increased EMG variability (r = 0.43, p = 0.034; Figure 5D). Pain, muscle soreness, functional limitation, age and gender for the overall sample and for the individuals who increased and decreased EMG variability at Day 4 are presented in Table 2. There was no relationship between map volume and EMG variability at Day 2 (rho = -0.16, p = 0.47) or Day 14 (r = -0.17, p = 0.43), nor between map volume and kinematic variability (all: p>0.51).

DISCUSSION

This study provides insight into motor adaptation, and the relationship between motor adaptation and corticomotor excitability, in response to progressively developing, sustained pain. At the group-level, EMG variability was decreased four days after pain onset and persisted at Day 14 despite resolution of pain. No changes were observed in kinematic data at the group level. However, there was substantial inter-individual variability in motor adaptation, with 36% and 52% of individuals displaying increased EMG and kinematic variability respectively, following four days of pain. A novel finding was that individuals who displayed increased EMG variability also displayed increased map volume, whereas individuals who decreased EMG variability displayed decreased map volume. These findings suggest a relationship between motor adaptation in pain and corticomotor excitability that is specific to the individual.

Previous studies have investigated motor adaptation in acute[4,24,55,91] or chronic pain[44,48,94], but evidence in the transition to sustained pain is limited. One previous NGF study evaluated the direction and variation of force during an isometric wrist extension task. That study found altered force direction, but not force variation, in radial-ulnar deviation following four days of pain[53]. The present study extends these findings by characterizing EMG and kinematic variability as pain persists over a similar time-frame. At the group level, we show reduced EMG variability, but no change in kinematic variability, following four days of pain. However, it was clear on visual inspection of the data that individuals adopted different motor strategies in response to pain. Sixteen (64%) participants reduced EMG variability at Day 4, while nine (36%) increased variability. Similarly, 52% of individuals increased (n=13), and 48% decreased (n=12), kinematic variability, explaining the insignificant group effect for this parameter.

The presence of inter-individual variability is consistent with the idea that motor adaptation to pain is not stereotypical, but specific to the individual[35,34] and with studies demonstrating individual-specific motor adaptation in acute experimental pain[33,70,78,87]. For example, when pain is induced in the back muscles via injection of hypertonic saline, 62% of individuals increase movement variability and 38% reduce variability[70]. Similarly, the current data suggest that when pain is sustained for four days, although all participants maintained task performance, the motor strategies used to achieve the task differed between individuals. Although both motor strategies (increased or decreased variability) may be successful at protecting the painful part in the short-term[34], individuals who adopt less variable motor patterns may be at greater risk of chronic pain as a result of stereotypical movement patterns that increase tissue loading[28,34,48,55,79]. Indeed, reduced motor variability in individuals with low back pain is considered a risk factor for the maintenance and reoccurrence of pain[22,55]. Further research is required to understand the trajectory of recovery following an episode of pain in those individuals that respond with an increase or decrease in motor variability.

Electromyography and kinematic variability failed to return to baseline at Day 14 in any subject despite resolution of pain. This was true regardless of whether individuals exhibited increased or decreased motor variability. This is in agreement with work demonstrating that changes in motor control with pain do not always resolve after pain has ceased[47,55,88]. For example, altered motor variability of postural strategies induced by acute experimental back pain persists after pain has resolved[55]. These findings support the notion that motor strategies adopted in the early stages of pain may not return to the original strategy following resolution of pain and could be a predisposing factor for the development of chronic pain[47,55,88].

One mechanism thought to contribute to motor adaptation in pain is altered corticomotor excitability[74,83,86,85]. Studies have shown increased corticomotor excitability when pain is sustained for four days[19,72]. In contrast, the present study found a decrease in corticomotor excitability. This discrepancy could be explained if the cortical strategy adopted in response to pain is not uniform across individuals. A recent study using the NGF model revealed individual differences in corticomotor excitability such that 60% of individuals displayed corticomotor depression and 40% displayed corticomotor facilitation[76]. Similar inter-individual variation was present in the current study with those who displayed reduced corticomotor excitability also displaying reduced motor variability and vice versa. This finding is consistent with motor learning studies that demonstrate a relationship between motor performance and corticomotor excitability[10-12]. These findings provide the first evidence of a link between corticomotor excitability and motor adaptation in response to pain that differs between individuals.

Why individuals adopt different motor and cortical strategies is not known. As pain intensity was not different between those who increased or decreased motor variability, these differences may relate to an individual's beliefs and attitudes towards pain, as well as their perception of threat[33,55,70]. For instance, those who perceive experimentally-induced back pain to be more threatening respond with a greater reduction in motor variability than those who perceive less threat[55]. Similarly, individuals who display high pain catastrophising and kinesiophobia to experimentally-induced back pain also display low variability in motor control strategies during trunk flexion-extension movements, whereas those who display low catastrophising and kinesiophobia display high variability[70]. Thus, one possibility is that individuals who perceived movement at the wrist to be highly threatening may have exerted tighter evaluative control over variability (leading to a decrease in motor variability and

corticomotor excitability), whereas those who perceived movement to be less threatening may have exerted less control over variability (leading to an increase in variability and corticomotor excitability).

Alternatively, experimentally induced nociception may have interfered with proprioceptive signaling and contributed to motor adaptation[52,84]. However, we consider this hypothesis less likely as although animal studies demonstrate excitation of high threshold mechanosensitive group IV muscle nociceptors with NGF[38,37], human studies show no effect of NGF on vibration sense or the jaw stretch reflex[81] and data on joint position sense collected as part of this protocol (published elsewhere) were unchanged over time[80]. It is also plausible that regions outside the sensorimotor cortex contributed to motor adaptation. For example, the cerebellum plays a key role in synchronization tasks[8,20] and studies have demonstrated activation of the cerebellum during the perception of pain[56]. Further research is needed to investigate the influence of activity in other brain regions on motor adaptation in pain.

The results of this study should be considered in light of several limitations. First, small training effects may have carried over between the pre-pain baseline sessions and the pain sessions. However, motor variability was stable across the two pain-free sessions suggesting that saturation in task variation occurred prior to NGF injection. Second, motor variability was assessed across the whole movement cycle using standard deviation, and other methods such as evaluating coordination variability between movement planes[31,64] or temporal structure of repetitive movements[66] were not included. Nonetheless, standard deviation is a valid measure of motor variability and has been widely used in pain studies[6,32,48,53-55,67]. Third, movement speed was controlled using a metronome,

meaning it was not possible for participants to adopt an alternative speed-accuracy strategy. Given the relevance of the speed-accuracy trade-off to motor skill performance[40], future studies should consider an individual's speed-accuracy strategy when investigating motor adaptation in pain. Fourth, single site excitability was calculated as the mean peak-to-peak MEP amplitude of five MEPs. While five MEPs have been shown to produce good between session reliability for calculating single site excitability [14], a higher number of stimuli (between 20-30) is required to optimise this assessment and produce excellent reliability[16,25]. Finally, re-learning of the task following a 10-day period with no task exposure may have contributed to the altered motor variability observed at Day 14. Previous literature has shown that motor skill performance degrades over time with extended delays in practice[23,71]. However, as a practice period was provided prior to each recorded trial, any influence of re-learning would likely have been minimal.

This study raises considerations for future research. As we were primarily interested in motor adaption as pain developed and peaked at Day 4, data were not collected between Days 4 and 14 and it is unknown if pain and corticomotor excitability follow a similar recovery trajectory. Future work should include a greater number of test sessions in the late stage of the model and incorporate daily pain diaries to ensure high temporal resolution of outcomes. It is also important to note that single-pulse TMS provides a measure of excitability along the entire corticomotor pathway. It is not possible to determine whether changes in corticomotor excitability occurred at spinal or cortical level using the current data. Similarly, intracortical inhibitory and facilitatory mechanisms which are known to be altered following four days of NGF-induced pain[72], were not evaluated. Further examination of spinal and intracortical mechanisms is needed. Lastly, it is conceivable that current or prior

engagement in musical or sporting activities could influence corticomotor excitability and motor adaptation in pain[63,89,41] and this information should be captured in future studies.

Conclusions

The motor strategy adopted in the transition to sustained pain is related to the motor cortical strategy, but the precise strategy differs between individuals. These findings are relevant given that altered motor variability is implicated in the persistence and recurrence of musculoskeletal pain[55,79].

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FIGURE LEGENDS

Figure 1. Experimental protocol. Participants attended five experimental sessions (Days -2, 0, 2, 4, and 14). At the beginning of each session, motor cortical maps were measured and motor variability assessed. On Days 0 and 2, injection of nerve growth factor (NGF) into the right ECRB muscle was performed immediately after completion of all outcome assessments. Pain intensity (NRS), muscle soreness (Likert scale), and functional limitation (patient rated tennis elbow evaluation, PRTEE) were recorded on Days 0, 2, 4, and 14.

Figure 2: Experimental set-up for radial/ulnar deviation task showing the position of the upper arm from the side view **(A)** and the view of board **(B)** positioned in front of each participant.

Figure 3: Representative example of ECRB EMG and wrist angle kinematics across a 45-repetition trial of the radial/ulnar deviation task. **A**. Radial/ulnar deviation angle (blue line) and ECRB EMG activity (red line). **B**. Forearm pronation/supination angle (orange line). **C**. Wrist flexion/extension angle (black line).

Figure 4. Mean (± SE, N = 25) pain NRS scores (**A**), Likert scores of muscle soreness (**B**), and functional limitation assessed by PRTEE (**C**) at each time-point (Days 0, 2, 4, and 14). * $P<0.05$ relative to Day 0 is illustrated.

Figure 5. Percent change in EMG (**A**) and kinematic (**B**) variability at Days 2, 4, and 14, normalised to baseline (Day 0). The black lines represent group data (mean ± SE; N = 25) and the grey lines individual data. Illustration of group mean (n=25, **C**) map volume obtained for the ECRB muscle at each time point (Days 0, 2, 4, and 14). Coordinates are referenced to the stimulation site that evoked the greatest motor evoked potential (centre grid reference in map) obtained for each individual. Maps for Days 2, 4, and 14 are normalised to the maximum MEP on Days 0. The coloured scale represents the proportion of the maximum MEP amplitude of Day 0. **D.** Scatter plot (N=25, percent change from baseline) showing the relationship between map volume and EMG variability at Day 4.

Table 1: Movement and corticomotor map measures (mean ± standard deviation) for Days -2 and 0.

	Day -2	Day 0
Kinematic variability (degree)	1.20 ± 0.45	1.25 ± 0.54
EMG variability (mV)	0.011 ± 0.006	0.012 ± 0.007
Performance accuracy (% accuracy)	96%	99%
Discrete peaks (number)	1.88 ± 0.93	1.84 ± 0.69
CoG latitude (cm)	5.79 ± 0.81	5.67 ± 0.75
CoG longitude (cm)	1.10 ± 0.68	1.14 ± 0.65
Map volume (mV)	8.04 ± 3.09	8.28 ± 2.58

EMG, electromyography; mV, millivolts; CoG, centre of gravity.

Table 2: Pain, muscle soreness, functional limitation, age and gender (mean ± standard deviation) for the overall sample and for the individuals

Variables	**Overall (n=25)**				**Increase EMG VAR (n=9)**				**Decrease EMG VAR (n=16)**			
	Day 0	**Day 2**	**Day 4**	**Day 14**	**Day 0**	**Day 2**	**Day 4**	**Day 14**	**Day 0**	**Day 2**	**Day 4**	**Day 14**
Pain	-	3.2±2.1	3.1±1.9	0.3±0.7	-	3.3±1.8	3.1±0.9	0.2±0.3	-	3.1±2.3	3.4±2.2	0.5±0.8
Muscle soreness	-	2.6±1.4	2.8±1.6	0.4±0.6	-	2.8±1.5	2.9±1.5	0.4±0.5	-	2.4±1.3	3.0±1.7	0.4±0.6
Functional limitation	-	9.8±6.7	11.8±8.2	0.9±2.4	-	9.9±6.2	11.5±5.9	0.3±0.3	-	9.7±7.5	11.6±5.2	0.5±0.6
Age (years)	23.9 ± 4.0				25.1±5.1				23.4±2.3			
Gender (male: female)	12:13				3:6				9:7			

who increased and decreased EMG variability at Day 4.

VAR, variability; EMG, electromyography; mV, millivolts; cm, centimetres