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RESEARCH ARTICLE

Physical Activity and the Brain

Motor cortex plasticity is greater in endurance-trained cyclists following acute exercise

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Abstract

Previous research using transcranial magnetic stimulation (TMS) has shown that plasticity within primary motor cortex (M1) is greater in people who undertake regular exercise, and a single session of aerobic exercise can increase M1 plasticity in untrained participants. This study aimed to examine the effect of an acute bout of exercise on M1 plasticity in endurance-trained (cyclists) and untrained individuals. Fourteen endurance-trained cyclists (mean ± SD; 23 ± 3.8 yr) and 14 untrained individuals (22 ± 1.8 yr) performed two experimental sessions. One session included an acute bout of high-intensity interval training (HIIT) exercise involving stationary cycling, whereas another session involved no-exercise (control). Following exercise (or control), I-wave periodicity repetitive TMS (iTMS) was used (1.5-ms interval, 180 pairs) to induce plasticity within M1. Motor evoked potentials (MEPs) induced by single and paired-pulse TMS over M1 were recorded from a hand muscle at baseline, after HIIT (or control) exercise and after iTMS. Corticospinal and intracortical excitability was not influenced by HIIT exercise in either group (all $P > 0.05$). There was an increase in MEP amplitude after iTMS, and this was greater after HIIT exercise (compared with control) for all subjects ($P < 0.001$). However, the magnitude of this response was larger in endurance cyclists compared with the untrained group ($P = 0.049$). These findings indicate that M1 plasticity induced by iTMS was greater in endurance-trained cyclists following HIIT. Prior history of exercise training is, therefore, an important consideration for understanding factors that contribute to exercise-induced plasticity.

NEW & NOTEWORTHY We use a novel form of repetitive transcranial magnetic stimulation to show that motor cortex plasticity is increased after acute exercise and that this effect is bolstered in endurance-trained cyclists. These findings indicate that participation in regular endurance exercise (involving lower limb muscles) has widespread effects on cortical plasticity (assessed in unexercised upper limb muscles) following acute lower-limb cycling exercise. It also highlights that exercise history is an important factor in exercise-induced cortical plasticity.

exercise; motor cortex; physical activity; plasticity; transcranial magnetic stimulation

INTRODUCTION

It is now commonly accepted that regular exercise has the potential to modulate the excitability of various cortical networks and promote neuroplasticity (1–3), which refers to the ability of the nervous system to modify the strength of its neural connections (4). It is likely that numerous physiological adaptations contribute to this upregulation of neuroplasticity with exercise. For example, long-term endurance exercise has been shown to promote cerebral angiogenesis and resultant blood flow (5, 6), as well as modulate circulating neurotrophins such as brain derived-neurotrophic factor (BDNF) (7), which have been shown to promote neuroplasticity (8). In addition, experimental evidence suggests that the increased plasticity associated with exercise may have functional benefits, such as greater cognitive function (9, 10) with long-term exercise, and improved motor skill learning (for review, see Ref. 11) and retention (12) following a bout of acute exercise. Despite these developments, it remains unclear if the effects of acute exercise on brain plasticity are modified in individuals who participate in regular endurance training.

Within the motor system, transcranial magnetic stimulation (TMS) has been regularly used to examine the role of acute exercise on excitability and plasticity of the corticospinal pathway and primary motor cortex (M1). Research suggests that acute exercise can modulate M1 and intracortical excitability in some cases; however, the effect is dependent on various factors including the intensity of acute exercise (13), and the participant's history of physical activity (2). Along with changes in M1 excitability, numerous studies have now demonstrated that a single bout of acute exercise increases the neuroplastic response to various plasticity-inducing paradigms involving repetitive TMS. For example, it

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was first shown that an acute bout of continuous exercise increases the long-term depression-like response to continuous theta burst stimulation (cTBS) (14), with subsequent studies showing that acute high-intensity interval training (HIIT) potentiates the long-term potentiation (LTP)-like response to both paired associative stimulation (PAS) (15) and intermittent theta burst stimulation (iTBS) (16). However, these neuroplastic responses after exercise are often quite variable (for review, see Ref. 17), suggesting that factors other than the type of acute exercise performed must also contribute to the neuroplastic response after acute exercise. One factor that may contribute to the variability in the plasticity response to acute exercise is the history of exercise or training in each participant. For example, baseline M1 excitability is altered in physically active individuals under some circumstances (1, 3, 18), and there is an increase in M1 excitability after exercise, but only in individuals with high physical activity levels (2). Similarly, M1 plasticity induced by PAS is greater in physically active individuals (3). However, it is unknown whether participation in long-term physical activity or exercise influences TMS-induced plasticity following an acute bout of exercise.

Therefore, the purpose of this study was to determine whether long-term endurance training modifies TMS-induced plasticity following a bout of acute aerobic exercise. The endurance-trained group consisted of highly trained cyclists with a history of endurance-based exercise, whereas an untrained control group consisted of individuals who did not participate in regular exercise. Each participant underwent an exercise protocol that involved HIIT, as this exercise modality is thought to be optimal for promoting M1 plasticity (17). As an alternative to the various plasticity inducing TMS protocols that have been implemented with exercise, such as iTBS (16, 19), cTBS (14), and PAS (15, 20), the present study utilized I-wave periodicity repetitive TMS (iTMS) to induce M1 plasticity following acute exercise. This intervention has been developed to target synaptic events within the intracortical circuits activated by TMS (21), producing robust effects (for review, see Ref. 22). Specifically, iTMS uses repetitive paired-pulse TMS at intervals of 1.5 ms, which coincides with the timing of indirect (I) waves present in the descending volley generated by a TMS pulse to M1 and targets specific interneuronal circuits that are known to be modulated with exercise (23). As a marker of M1 plasticity, we measured the change in corticospinal excitability and M1 intracortical facilitation and inhibition after HIIT exercise and after iTMS. Given that acute exercise (14–16) and regular physical activity (3) are known to improve M1 plasticity, it was hypothesized that iTMS-induced M1 plasticity after HIIT would be greater in endurance-trained participants.

METHODS

Twenty-eight participants were recruited from the university and the broader community to participate in the current study. This study included two participant groups that were recruited based on their specific training history and cumulative physical activity levels, which were assessed using the International Physical Activity Questionnaire (IPAQ) short form (24). Sample size calculations were performed using G*Power 3.1.9.7 software and were based on the expected modulation of motor evoked potentials (MEPs) after iTMS (25) and group differences in M1 plasticity between physically active and untrained participants (3). This analysis revealed that 13 untrained and 13 endurance-trained participants were required to detect a significant difference in MEP modulation between groups after iTMS, with at least 80% power and alpha error of 0.05. An endurance-trained group, referred to as *ET* group, consisted of 14 highly trained cyclists (mean age ± SD: 23 ± 3.8 yr, 3 female) who each had a total physical activity level of greater than 3,000 MET minutes of weekly exercise (14, 19), including 6 or more hours of cycling but no formal upper body exercise. An untrained group, referred to as *UT* group, included 14 (22 ± 1.8 yr, 4 female) inactive individuals who had a total physical activity level of no more than 1,500 MET minutes per week (24) and did not participate in any regular aerobic training. In addition, a physical activity readiness questionnaire (PAR-Q) was used to assess suitability to complete exercise with acceptable risk (26). Exclusion criteria for all participants included a history of concussion, neurological disease, or ongoing use of psychoactive medication (antidepressants, sedatives, etc.). All experimentation was approved by the University of Adelaide Human Research Ethics Committee and conducted according to the Declaration of Helsinki. Each participant provided written, informed consent before inclusion in the study.

Experimental Arrangement and Procedures

Each participant attended two experimental sessions. One exercise session, referred to as the *HIIT* session, included a 20-min bout of HIIT cycling. A separate session utilized a control exercise protocol in place of the cycling exercise and is referred to as the *control* session. The experimental protocol and all TMS measurements were consistent between the two sessions, with the only difference being the exercise/no exercise control (Fig. 1). These sessions were conducted a minimum of 7 and a maximum of 30 days apart. To control for the elevation in cortisol after waking, which is known to inhibit plasticity induction (27), all experiments were commenced after 11:00 AM. Furthermore, to avoid exercise-related confounds, all participants were instructed not to complete any exercise on the day of testing, before the experimental session. A Polar M400 heart rate monitor (polar electro) was used for both sessions to assess baseline resting heart rate (obtained after 5 min of quiet sitting) and to monitor heart rate throughout the exercise or control block. For TMS measurements, subjects were seated in a comfortable chair, with feet placed flat on the floor and the right hand relaxed on a benchtop. Surface electromyography (EMG) was recorded from the first dorsal interosseous (FDI) muscle of the right hand using two Ag-AgCl electrodes, in accordance with the European recommendations of surface electromyography (28). An earth strap was fitted around the wrist to ground the electrodes. EMG signals were amplified (300×) and band-pass filtered (20 Hz high pass, 1 kHz low pass) using a CED1902 signal conditioner (Cambridge Electronic Design, Cambridge, UK) before being digitized at 2 kHz using a CED1401 interface. Recordings were stored on a computer for offline analysis.

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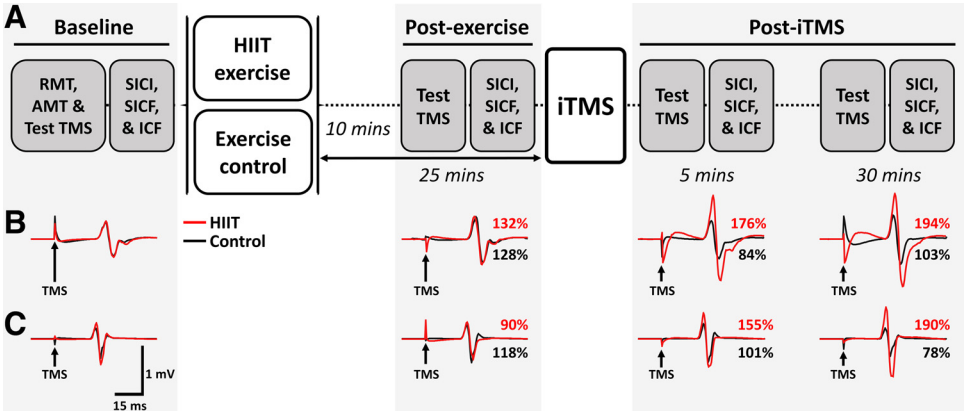

Figure 1. Experimental protocol. *A*: session protocol. Traces display test MEP responses recorded in a single endurance-trained (*B*) and untrained (*C*) participant at the four TMS testing time points. Black traces display mean MEP waveforms from the control session and red traces depict MEP waveforms recorded during the HIIT session. Inset percentage values at the postexercise time point represent the mean MEP amplitude relative to baseline for each condition. Inset percentage values at the post-iTMS time points represent the mean MEP amplitudes relative to the postexercise time point, for each condition. HIIT, high-intensity interval training; ICF, intracortical facilitation; iTMS, I-wave periodicity repetitive TMS; MEP, motor evoked potential; SICF, short-interval intracortical facilitation; SICI, short-interval intracortical inhibition; TMS, transcranial magnetic stimulation.

Transcranial Magnetic Stimulation

In both sessions, transcranial magnetic stimulation (TMS) was applied to the left M1 using a figure-of-eight coil connected to two Magstim 200^2 magnetic stimulators through a Bistim module (Magstim, Dyfed, UK). The coil was positioned to induce a posterior-anterior (PA) current within M1 by directing the handle posterior-laterally at an angle of ~45° to the midline. The coil was held tangentially to the scalp over the optimal location for producing MEPs in the target muscle. This location was marked on the scalp for reference and checked throughout the experiment. TMS was delivered at 0.2 Hz with a 10% variation between trials to avoid anticipation of the stimulus.

Single-pulse TMS measures consisted of resting motor threshold (RMT), active motor threshold (AMT), and test TMS. RMT was assessed at the beginning of each session, which was defined as the minimum stimulus intensity required to produce an MEP with an amplitude of ≥50 μV in at least 5 out of 10 consecutive trials in the relaxed target muscle. For AMT, participants were instructed to maintain a contraction intensity of 10% maximal voluntary contraction (MVC), with visual feedback of force provided on an oscilloscope. AMT was subsequently measured and defined as the minimum TMS intensity required to produce an MEP with an amplitude of >200 μV in at least 5 out of 10 consecutive trials. A test TMS intensity was defined as the stimulus intensity required to produce a peak-to-peak MEP amplitude of ~1 mV (range, 0.5–1.5 mV) when averaged over 20 trials, and this same intensity was used at each time point to examine the change in MEP amplitude after exercise and after iTMS (relative to baseline).

Paired-pulse TMS was used to assess intracortical excitability with measures of short-interval intracortical inhibition (SICI), short-interval intracortical facilitation (SICF), and intracortical facilitation (ICF). Measures of SICI utilized a subthreshold conditioning pulse set at 80% of AMT (29, 30) and an interstimulus interval (ISI) between conditioning and test stimuli of 2 ms (29). SICF was obtained with a subthreshold conditioning intensity of 90% RMT, which was applied 1.5 ms following the test pulse (2, 31). ICF was assessed using a subthreshold conditioning stimulus of 80% RMT applied 10 ms before the test pulse (32). Conditioning intensities for all paired-pulse TMS measures remained the same at each time point (33), as motor threshold is not altered with acute exercise (19). However, the test TMS intensity for paired-pulse TMS at each time point (after exercise and iTMS) was adjusted to produce a mean MEP response that was within 30% of the baseline mean (3, 16). To achieve this, a single-pulse test block of 20 trials was recorded at the baseline test TMS intensity. If the mean MEP amplitude was not within 30% of the baseline mean, TMS intensity was adjusted and a subsequent block of 20 trials was completed to ensure the intensity was adjusted appropriately (within 30% of the baseline mean), and this adjusted intensity was used for the paired-pulse responses.

At baseline, a single-pulse test block (n = 20 trials) was applied to measure corticospinal excitability at the predetermined test TMS intensity. Subsequently, a paired-pulse test block was completed to assess measurements of SICI, SICF, and ICF. These test blocks comprised 15 test TMS stimuli, as well as 15 trials of each paired-pulse condition (SICI, SICF, and ICF) in a pseudorandomized order. Following baseline TMS measurements, participants completed either the HIIT or control exercise block. Both the single- and paired-pulse test blocks were then repeated 10 min after the completion of exercise (or control), before iTMS was administered (25 min after exercise completion). Single- and paired-pulse TMS blocks were repeated at 5 and 30 min following iTMS (Fig. 1).

Exercise Protocol

For the exercise session, participants pedaled on a stationary cycling ergometer (Wattbike, UK) for 20 min in a HIIT protocol (33). Exercise intensity was personalized using each individual's age-predicted maximum heart rate (220 minus age; HR_{max}) and resting heart rate (RHR), which were used to calculate heart rate reserve (HRR = HR_{max} − RHR). The HIIT cycling involved 3 min at low intensity (RHR + 50% HRR) followed by 2 min of high-intensity cycling (RHR + 90% HRR), repeated four times for a total duration of 20 min (33).

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At the completion of the exercise block, participants continued to pedal with low resistance for 2 min to cool down before sitting quietly for a further 8 min before application of TMS. During the control session, participants sat on the stationary bicycle in the same position as during the exercise session, but were instructed not to pedal. During exercise and control blocks, participants were instructed to keep their hands relaxed on the handlebars to avoid activation of the FDI muscle. Compliance was assessed by recording surface EMG bilaterally from FDI at 1,000 Hz using a Mega ME6000 biomonitor (Mega Electronics, Finland), with visual feedback provided to the participants. To normalize the EMG to maximum, participants completed two MVCs of each hand immediately before the exercise. Exercise EMG measures were stored on a CompactFlash for later analysis through Megawin software (Mega electronics, Finland) and custom MATLAB scripts. During HIIT and control exercise blocks, data were recorded for heart rate and average power, as well as a self-reported rating of perceived exertion (34), and an affect score of feeling and emotion (35). These measures were obtained at the end of each low- or high-intensity block (or corresponding time points for the control session).

I-Wave Periodicity Repetitive TMS

Plasticity within M1 circuits was induced using I-wave periodicity repetitive TMS (iTMS), which consisted of 180 pairs of stimuli (ISI of 1.5 ms) applied every 5 s for a period of 15 min (21). The intensity of TMS was the same for each stimulus in the pair, and was set to produce a paired-pulse MEP of 0.5–1.0 mV at the start of the block (21). During the intervention, participants were instructed to observe their EMG activity displayed on an oscilloscope and ensure consistent muscle relaxation. Small ice packs were applied to the TMS coil before the intervention to alleviate coil heating during iTMS.

Data Analysis

During offline analysis of MEP data from the resting session, trials containing EMG activity >20 μV (peak-to-peak amplitude) in the 100 ms before TMS application were discarded. MEPs were measured peak-to-peak for each trial and expressed in mV. To assess the influence of intracortical networks on MEP amplitude, individual MEPs recorded in response to paired-pulse TMS were expressed as a percentage of the mean test TMS amplitude. Accordingly, normalized values greater than 100% reflect facilitation, whereas normalized values of less than 100% reflect inhibition. For these paired-pulse measures, it was determined a priori that only participants who displayed the expected response at baseline (<100% of test MEP for SICI and >100% test MEP for SICF and ICF) were included in analyses, which is in line with previous studies (2, 36). This was based on the rationale that it would be difficult to interpret the modulation of a response (with exercise or iTMS) if the expected response was not detected at baseline. Subsequent statistical analysis indicated that the exclusion of these participants (who did not demonstrate a response at baseline) did not alter the main outcomes of the paired-pulse TMS data after exercise or iTMS.

To identify exercise-induced changes in both corticospinal and intracortical excitability, normalized MEPs from single- and paired-pulse TMS blocks recorded after exercise were expressed as a percentage of the mean values from baseline. To identify iTMS-induced changes in excitability, normalized MEPs from single- and paired-pulse TMS blocks recorded 5- and 30-min post-iTMS were expressed as a percentage of the mean values for the same participant obtained at the postexercise time point. To ensure that this normalization approach produced robust outcomes, we performed an additional analysis where the absolute values for single (MEP) and paired-pulse TMS (SICI, SICF, and ICF) in each participant was subtracted from the baseline measurements at each time point (postexercise minus baseline; post-iTMS minus postexercise). Statistical analysis of these data using the subtraction approach (data not shown) resulted in similar outcomes to the normalization approach used here, supporting the main conclusions of the study. This suggests that the normalization approach used here provides an accurate reflection of the main study outcomes.

To quantify FDI muscle activity during the exercise and control (no exercise) sessions, EMG recorded was normalized to maximum (EMG_{max}), which was obtained from the MVCs performed before exercise. Activity greater than 5% of EMG_{max} was categorized as a "burst" of activity, with the total number of bursts and mean burst amplitude recorded for each session and group. Furthermore, cumulative burst time was expressed relative to the total recording time and referred to as total active time. As a measure of the net activity of the FDI during exercise, the intensity of the total burst activity (%MVC) was expressed relative to time (h) as the summed area of activity (%MVC/h). For exercise-related measures, exercise intensity was expressed as %HRR (above RHR), and power was normalized to body weight and expressed as watts:kg. Exercise data [power:weight, exercise intensity (%HRR), rating of perceived exertion (RPE), and affect scores] from the HIIT block were grouped into low- and high-intensity categories. Mean values were expressed for each group for the control session and for group and intensity separately for the HIIT session.

Statistical Analysis

The normality of data was assessed using Shapiro–Wilk tests, and log transformations were applied when this test indicated deviation from a normal distribution. All data are displayed in the original form (nontransformed) for clarity. Unpaired *t* tests were used to compare mean weekly exercise load (MET minutes) between groups (ET, UT). Baseline single-pulse measures of corticospinal excitability (test TMS) and paired-pulse measures of intracortical excitability (SICI, SICF, and ICF) were compared between groups and sessions (HIIT, control) using separate two-factor repeated measures linear mixed model (LMM_{RM}) analyses.

For analysis of EMG activity recorded during the exercise blocks, the number of bursts, mean burst amplitude, active time and summed area of activity were compared between groups and sessions using separate two-factor LMM_{RM} analyses. Furthermore, nontransformed data were used within Spearman's correlation analyses (which are known to be particularly robust against outliers, 37) to determine whether there was an association between activation of FDI during HIIT and the exercise- and iTMS-induced modulation of

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corticospinal excitability. Separate analyses were run for each of the exercise EMG variables (number of bursts, mean burst amplitude, active time, and summed area of activity) against the normalized TMS measures of excitability recorded after exercise and iTMS. These correlation analyses were undertaken with both groups combined and separately. Outcomes from the HIIT exercise block (power: weight, exercise intensity [%HRR], RPE, and affect score) were compared independently between group and intensity (low, high) using a two-factor LMM. To compare overall outcomes between the HIIT exercise and control blocks, measures of exercise intensity (%HRR), RPE, and affect score were compared between groups and sessions in separate two-factor LMM_{RM} analyses.

The change in corticospinal and intracortical excitability with exercise and iTMS were assessed with separate one-sample t tests with Bonferroni correction, whereby normalized MEP responses from postexercise and post-iTMS time points were compared with a 100% test value for each condition (group, session). Furthermore, normalized TMS measures (test TMS, SICI, SICF, ICF) from the postexercise time point were compared between groups and sessions using separate two-factor LMM_{RM} analyses. TMS measures after iTMS (normalized to postexercise) were compared between groups, sessions, and time points (5 post, 30 post) using a three-factor LMM_{RM}. Raw test TMS amplitudes from the paired-pulse test blocks were also compared between group, session, and time point (baseline, postexercise, 5 post- and 30 post-iTMS) using a three-factor LMM_{RM}. Spearman's analyses were used to assess correlations between normalized TMS measures (test TMS, SICI, SICF, ICF) recorded after the exercise block and the change in test MEP amplitude with iTMS, as well as to compare each of the paired-pulse measures recorded after iTMS (normalized to postexercise) to the test MEP recorded after iTMS.

For all models, subject was included as a random effect, and significant main effects and interactions were further investigated using custom contrasts with Bonferroni correction. As the aims of this study were concerned with the effect of regular endurance training on plasticity, only interactions in which group was a factor were investigated. For all significant effects and interactions of LMM analyses, the estimated mean difference (EMD) and associated 95% confidence interval from post hoc pairwise comparisons have been presented as nonstandardized measures of effect size (38). EMDs were exponentiated from the log values derived from individual pairwise comparisons of estimated marginal means and, therefore, represent a ratio of the means being compared. Tables and figures display mean values of all participants included in the corresponding analysis. Data are displayed as mean ± 95% confidence interval unless stated otherwise. For clarity, 5- and 30-min time points are grouped in figures.

RESULTS

All participants completed the study in full and without any adverse effects. The IPAQ showed a significantly greater weekly exercise load of 5,850 MET minutes for the ET group compared with 1,139 MET minutes for the UT group ($P < 0.001$). Furthermore, based on the self-reported activity levels within the IPAQ, the ET group accumulated 54% of their total weekly MET minutes through vigorous-intensity exercise (predominantly endurance-based cycling), compared with only 12% for the UT group, which was mostly obtained through normal daily activities (i.e., lifting/carrying).

Baseline Measures of Corticospinal and M1 Intracortical Excitability

Table 1 displays the baseline TMS data for each participant group. LMM_{RM} revealed no differences in RMT, AMT, or test TMS intensity and no difference in mean test TMS amplitude between groups or sessions (all $P > 0.05$). For paired-pulse TMS, participants were excluded from the analysis if they did not show the expected response at baseline, resulting in 11 ET participants for SICI, 13 ET participants for SICF, and 10 ET and 10 UT participants for ICF from a total of 14 participants in each group. Baseline measures of SICI and ICF did not vary between groups or sessions and no significant interactions were obtained (all $P > 0.05$). Although baseline SICF

Table 1. *Baseline TMS data*

	Control		HIIT	
	ET	UT	ET	UT
TMS intensity RMT, % MSO	41.5 (37.9, 45.1)	41.5 (38.0, 45.0)	41.1 (38.1, 44.1)	40.7 (37.1, 44.3)
TMS intensity AMT, % MSO	34.3 (31.4, 37.2)	35.3 (31.9, 38.6)	35.3 (32.6, 38.0)	34.8 (31.6, 37.9)
TMS intensity test, % MSO	51.4 (44.9, 57.9)	49.8 (44.8, 54.8)	51.8 (46.6, 57.0)	49.6 (44.4, 54.7)
Test response, mV	0.8 (0.7, 0.9)	0.9 (0.8, 1.0)	0.9 (0.8, 1.0)	0.9 (0.8, 1.0)
SICI, % test	48.1 (41.2, 55.1)	39.6 (34.1, 45.0)	45.3 (39.7, 50.9)	47.0 (41.2, 52.8)
SICF, % test	219.8 (201.1, 238.6)	207.9 (190.0, 225.8)	178.2* (160.5, 196.0)	194.8 (180.6, 209.0)
ICF, % test	170.4 (151.9, 189.0)	177.5 (158.8, 196.2)	218.0 (182.5, 253.6)	149.3 (136.7, 162.0)

Data represent mean (95% CI). *$P < 0.005$ compared with control session (ET group). AMT, active motor threshold; CI, confidence interval; ET, endurance trained; ICF, intracortical facilitation; MSO, maximum stimulator output; RMT, resting motor threshold; SICF, short-interval intracortical facilitation; TMS, transcranial magnetic stimulation; UT, untrained.

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Table 2. *Exercise EMG data*

	Control			HIIT		
	ET	UT	All	ET	UT	All
Number of bursts	52.2 (0.3, 104.2)	76.2 (−13.3, 165.6)	63.7 (16.8, 110.7)	145.7 (73.1, 218.2)	197.0 (96.2, 297.8)	171.3* (112.9, 229.8)
Mean burst amplitude, %MVC	8.3 (5.5, 11.2)	9.8 (5.3, 14.3)	9.0 (6.6, 11.4)	10.6 (8.3, 13.0)	10.8 (9.0, 12.6)	10.7 (9.4, 12.1)
Active time, % total time	2.2 (−0.1, 4.6)	3.0 (−0.7, 6.6)	2.6 (0.6, 4.6)	12.2 (3.2, 21.3)	15.1 (5.7, 24.6)	13.7* (7.6, 19.8)
Summed area of activity, %MVC/h	0.2 (0.1, 0.4)	1.4 (−1.3, 4.1)	0.8 (−0.4, 2.0)	1.4 (0.2, 2.6)	1.9 (0.5, 3.2)	1.6* (0.8, 2.5)

Data represent mean (95% CI). *$P < 0.005$ compared with control session. EMG, electromyography; CI, confidence interval; ET, endurance trained; HIIT, high-intensity interval training; MVC, maximal voluntary contraction; UT, untrained.

was not different between groups ($F_{1, 25} = 0.03$, $P = 0.9$), it was significantly greater in the control compared with the HIIT session (EMD = 112%, 95% CI [103, 123], $F_{1, 208} = 6.8$, $P = 0.01$). Differences between sessions also varied by group (group × session interaction, $F_{1, 208} = 5.8$, $P = 0.02$), with post hoc tests revealing that for the ET group, baseline SICF was greater in the control session compared with the HIIT exercise session (EMD = 125%, 95% CI [110, 142], $P < 0.001$), whereas there was no difference between sessions for the UT group (EMD = 101%, 95% CI [89, 114], $P = 0.9$).

HIIT Exercise

For recordings of FDI muscle activity during exercise, LMM_{RM} revealed no difference between groups for number of EMG bursts, mean burst amplitude, total active time, or summed area of activity (all $P > 0.6$). Furthermore, values did not differ between sessions for mean burst amplitude ($P = 0.7$) but were greater in the HIIT exercise block compared with control for number of bursts, active time and summed area of activity (all $P < 0.005$; Table 2).

Table 3 displays the outcomes of HIIT exercise and control sessions (power to weight ratio, exercise intensity, RPE, and affect score). LMM analysis of data from the HIIT session revealed significantly greater power to weight ratio for the ET group compared with the UT group (EMD = 222%, 95% CI [183, 270], $F_{1, 26} = 70$, $P < 0.001$), and during high- compared with low-intensity phases of HIIT (EMD = 380%, 95% CI [343, 422], $F_{1, 64} = 653$, $P < 0.001$), but there was no interaction

Table 3. *Exercise data for HIIT and control sessions*

	Control	HIIT		
	Rest	Low Intensity	High Intensity	Total
Power to weight ratio, watts:kg				
ET	0.0 (0.0)	1.4 (1.2, 1.5)	4.6 (4.4, 4.8)	3.0 (2.7, 3.3)*
UT	0.0 (0.0)	0.7 (0.6, 0.8)	2.3 (2.1, 2.4)	1.5 (1.3, 1.6)
All	0.0 (0.0)	1.0 (0.9, 1.1)	3.4 (3.2, 3.7) +	2.2 (2.0, 2.4)
Exercise intensity, %HRR				
ET	13.2 (11.7, 14.6)	51.3 (50.0, 52.6)	88.5 (87.2, 89.8) +	69.9 (66.3, 73.5)
UT	14.1 (12.2, 15.9)	53.5 (51.8, 55.2)	85.9 (83.2, 88.5) +	69.7 (66.3, 73.1)
All	13.6 (12.5, 14.8)	52.4 (51.3, 53.5)	87.2 (85.7, 88.7) +	69.8 (67.3, 72.3)^
Rating of perceived exertion				
ET	6.6 (6.6, 6.7)	9.4 (8.8, 10.0)*	16.6 (16.2, 17.0) +	13.0 (12.2, 13.8)^
UT	6.4 (6.3, 6.5)	10.5 (9.8, 11.2)	15.9 (15.4, 16.5) +	13.2 (12.6, 13.9)^
All	6.5 (6.5, 6.6)	9.9 (9.5, 10.4)	16.3 (15.9, 16.6) +	13.1 (12.6, 13.6)^
Affect score				
ET	4.4 (4.2, 4.5)	2.9 (2.5, 3.4)	1.7 (1.3, 2.2)	2.3 (2.0, 2.7)*
UT	3.7 (3.5, 4.0)	2.0 (1.5, 2.5)	0.3 (−0.3, 1.0)	1.2 (0.7, 1.6)
All	4.0 (3.9, 4.2)	2.5 (2.1, 2.8)	1.0 (0.6, 1.4) +	1.7 (1.5, 2.0)^

Data represent mean (95% CI). For HIIT sessions, data are grouped by intensity, with means for each displayed separately, as well as a "total" column displaying values grouped by intensity. *$P < 0.05$ compared with UT group; $^{+}P < 0.001$ compared with the low intensity; ^$P < 0.001$ compared with control session. CI, confidence interval; ET, endurance trained; HIIT, high-intensity interval training; HRR, heart rate reserve; UT, untrained.

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between factors ($F_{1, 64}$ = 1.3, P = 0.3). For exercise intensity (% HRR), values did not differ between groups ($F_{1, 26}$ = 0.001, $P > 0.9$) but were significantly greater during high- compared with low-intensity phases of HIIT (EMD = 166%, 95% CI [163, 169], $F_{1, 82}$ = 3,156, $P < 0.001$). Differences between high and low intensities also varied between groups (exercise intensity × group interaction, $F_{1, 82}$ = 18, $P < 0.001$). Post hoc tests revealed that exercise intensity (%HRR) was greater for high- compared with low-intensity phases of exercise for each group (both $P < 0.001$). Although RPE was not different between groups ($F_{1, 26}$ = 0.5, P = 0.5), it was greater in the high- compared with the low-intensity phase of HIIT (EMD = 167%, 95% CI [160, 175], $F_{1, 65}$ = 533, $P < 0.001$). Differences between RPE also varied by group ($F_{1, 65}$ = 12.1, P = 0.001), with post hoc tests revealing that scores in the low-intensity phase were reduced in the ET group compared with the UT group (EMD = 90%, 95% CI [81, 99], P = 0.04) but were not different between groups for the high-intensity phase (EMD = 105%, 95% CI [95, 116], P = 0.3). Furthermore, RPE values were greater in the high- compared with the low-intensity phase of HIIT for each group (both $P < 0.001$). For affect scores, values were significantly greater for the ET group compared with the UT group (EMD = 112%, 95% CI [100, 123], $F_{1, 26}$ = 4.4, $P < 0.05$), and for the low- compared with high-intensity phase of HIIT (EMD = 114%, 95% CI [112, 116], $F_{1, 88}$ = 83, $P < 0.001$), but there was no interaction between factors ($F_{1, 88}$ = 2.4, P = 0.1). LMM_{RM} comparisons between HIIT and control exercise blocks revealed significantly greater values in the HIIT session compared with the control session for measures of exercise intensity (EMD = 569%, 95% CI [490, 662], $F_{1, 135}$ = 517, $P < 0.001$) and RPE (EMD = 192%, 95% CI [187, 196], $F_{1, 241}$ = 2,769, $P < 0.001$), whereas affect score was lower in the HIIT session compared with the control session (EMD = 79%, 95% CI [77, 84], $F_{1, 111}$ = 102, $P < 0.001$). For RPE, differences between sessions also varied by group, with post hoc tests revealing that RPE values were significantly greater for the HIIT session in ET and UT groups (both $P < 0.001$).

Corticospinal and M1 Intracortical Excitability Following HIIT Exercise

Figure 2 displays mean MEP amplitudes and individual participant responses from TMS measures of corticospinal excitability recorded at the postexercise time point (expressed relative to baseline). MEP amplitudes did not differ from baseline for either group after HIIT or control (one-sample t tests; all $P > 0.05$). LMM_{RM} analysis for normalized MEP amplitudes revealed no difference between groups ($F_{1, 26}$ = 1.1, P = 0.3); however, values were significantly lower for the HIIT session compared with the control session (EMD = 72%, 95% CI [62, 83], $F_{1, 266}$ = 20, $P < 0.001$; Fig. 2A). Differences in normalized MEP amplitude between sessions also varied by group (group × session interaction, $F_{1, 266}$ = 8.7, P = 0.003), with post hoc tests showing that the normalized MEP amplitude was lower in the HIIT session compared with the control session (EMD = 58%, 95% CI [47, 71], P = 0.001) in the ET group, whereas there was no difference between sessions for the UT group (EMD = 89%, 95% CI [73, 109], P = 0.3). Furthermore, ET participants displayed greater normalized MEP amplitude compared with the UT group in the control session (EMD = 148%, 95% CI [103, 213], P = 0.04), but there was no difference between groups for the HIIT session (EMD = 96%, 95% CI [67, 138], P = 0.8).

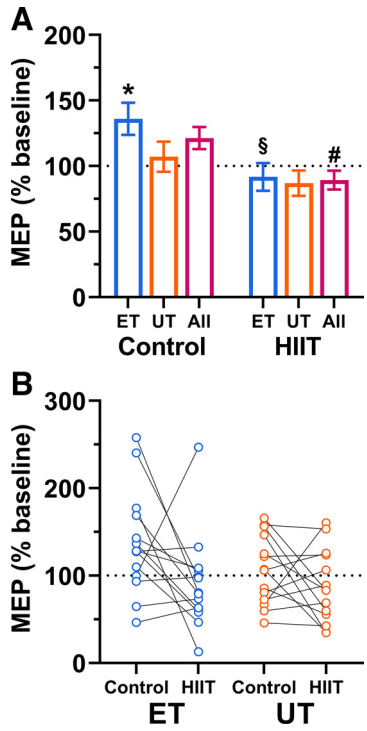

Figure 2. Effect of high-intensity interval training (HIIT) and control (no exercise) on corticospinal excitability assessed with single-pulse TMS. Data were recorded at the postexercise time point and represent mean MEP amplitude (*A*) and individual subject means matched between sessions (*B*; ET n = 14, UT n = 14) after exercise relative to baseline. Data points above 100% indicate greater test MEP amplitude relative to baseline. Repeated measures linear mixed model analysis, *P = 0.04 compared with UT group (control session); §P = 0.001 compared with control session (ET group); #$P < 0.001$ compared with control session (groups combined). ET, endurance-trained; MEP, motor evoked potential; UT, untrained.

Paired-pulse TMS measures of SICI, SICF, and ICF did not differ from baseline in either group after the HIIT or control sessions (one-sample t tests; all $P > 0.05$; Fig. 3). LMM_{RM} analysis between groups and sessions showed no significant effects or interactions for SICI or SICF (all $P > 0.05$). For ICF, normalized values were not different between groups >($F_{1, 18}$ = 0.3, P = 0.6) or sessions ($F_{1, 158}$ = 0.04, P = 0.8); however, a significant interaction between group and session was present ($F_{1, 158}$ = 9.2, P = 0.003; Fig. 3C). Post hoc tests revealed that normalized ICF values were significantly lower in the HIIT session compared with the control session in the ET group (EMD = 82%, 95% CI [68, 100], $P < 0.05$); however, normalized ICF was greater in the HIIT session compared with the control session in the UT group (EMD = 124%, 95% CI [103, 151], P = 0.02).

Corticospinal and M1 Intracortical Excitability Following iTMS

The effect of iTMS on corticospinal excitability is shown in Fig. 4, where the MEP amplitudes after iTMS have been normalized to the MEP amplitudes obtained after the HIIT or control session (referred to as the postexercise time point). When compared with postexercise, one-sample t

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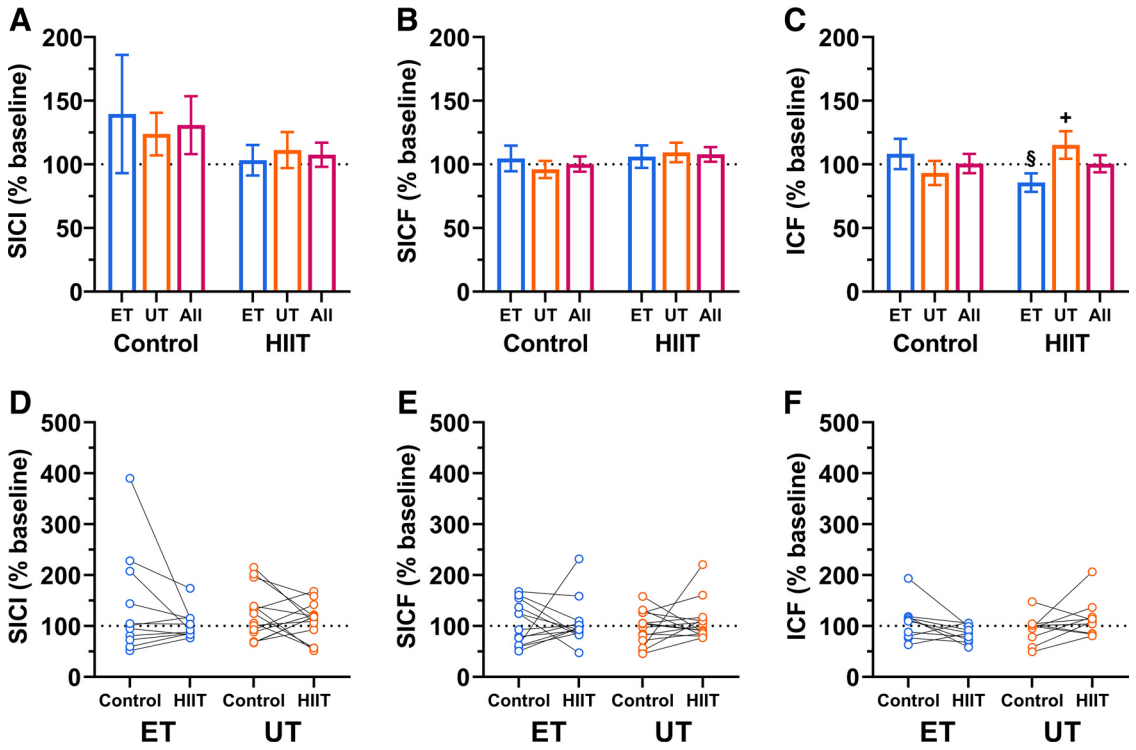

Figure 3. Effect of high-intensity interval training (HIIT) and control (no exercise) on intracortical excitability assessed with paired-pulse TMS. Data were recorded at the postexercise time point and represent mean response amplitudes (*top row*) and individual subject means matched between sessions (*bottom row*) after exercise relative to baseline for SICI (*A* and *D*; ET $n = 11$, UT $n = 14$), SICF (*B* and *E*; ET $n = 13$, UT $n = 14$), and ICF (*C* and *F*; ET $n = 10$, UT $n = 10$). Data points above 100% indicate a reduction of SICI (*A* and *D*) and greater SICF (*B* and *E*) and ICF (*C* and *F*), compared with baseline. Repeated measures linear mixed model analyses, §$P < 0.05$ compared with control session (ET group); +$P = 0.02$ compared with control session (UT group). ET, endurance-trained; ICF, intracortical facilitation; SICF, short-interval intracortical facilitation; TMS, transcranial magnetic stimulation; UT, untrained.

tests showed that test MEP amplitudes were not significantly modulated in the control session for either group (both $P > 0.05$). However, test MEP amplitudes increased in both the ET ($P = 0.007$) and UT ($P = 0.004$) group in the HIIT session. Furthermore, with groups combined, test MEP amplitudes increased with iTMS in both the control ($P = 0.004$) and HIIT ($P < 0.001$) sessions. LMM_{RM} analysis of test MEP amplitude revealed there was no difference between groups ($F_{1,\ 26} = 1.2$, $P = 0.3$); however, normalized MEP amplitudes after iTMS were significantly greater in the HIIT exercise session compared with the control session (EMD = 133%, 95% CI [119, 148], $F_{1,\ 484} = 26.3$, $P < 0.001$; Fig. 4*A*), and at the 30-min time point compared with 5-min post-iTMS (EMD = 114%, 95% CI [102, 127], $F_{1,\ 504} = 5.6$, $P = 0.02$). Differences between sessions also varied by group (group × session interaction, $F_{1,\ 484} = 6.9$, $P = 0.009$), with post hoc tests revealing that normalized MEP amplitudes after iTMS were greater in the ET group compared with UT group after HIIT exercise (EMD = 134%, 95% CI [100, 179], $P < 0.05$). However, there was no difference between groups for the control session (EMD = 100%, 95% CI [75, 134], $P > 0.9$). In addition, normalized MEP amplitudes for the ET group were greater in the HIIT session compared with the control session (EMD = 153%, 95% CI [132, 179], $P < 0.001$), whereas there was no difference between sessions for the UT group (EMD = 115%, 95% CI [98, 134], $P = 0.07$).

The effect of iTMS on intracortical excitability assessed with paired-pulse TMS is shown in Fig. 5. LMM_{RM} revealed that test MEP amplitudes recorded from the paired-pulse blocks did not vary between groups ($F_{1,\ 26} = 0.1$, $P = 0.8$) or sessions ($F_{1,\ 779} = 1.3$, $P = 0.2$). However, they differed over time point ($F_{3,\ 914} = 5.8$, $P = 0.001$), being greater than baseline (mean = 1.07 mV) at 5 post (mean = 1.33 mV, EMD = 117%, 95% CI [102, 135], $P = 0.01$) and 30 post (mean = 1.31 mV, EMD = 123%, 95% CI [107, 142], $P < 0.001$). When examining the change in paired-pulse TMS responses after iTMS (relative to postexercise), there were no significant changes in SICI, SICF, or ICF (one-sample t tests; all $P > 0.05$). LMM_{RM} analysis of SICI after iTMS revealed no significant effects of group ($F_{1,\ 23} = 0.2$, $P = 0.7$), session ($F_{1,\ 385} = 0.008$, $P > 0.9$), or time point ($F_{1,\ 449} = 1.2$, $P = 0.3$), and no significant interactions were present (all $P > 0.09$). For SICF, there was no effect of group ($F_{1,\ 25} = 0.2$, $P = 0.6$) or time point ($F_{1,\ 432} = 1.7$, $P = 0.2$); however, the change in SICF with iTMS was greater in the control session compared with the HIIT exercise session (EMD = 93%, 95% CI [86, 100], $F_{1,\ 339} = 4.4$, $P = 0.04$; Fig. 5*B*). Differences between sessions also varied by group (group × session interaction, $F_{1,\ 339} = 5.3$, $P = 0.02$) with post hoc tests revealing that for the UT group, the change in SICF after iTMS was greater in the control session compared with the HIIT session (EMD = 85%, 95% CI [77, 94], $P = 0.002$), whereas there was no difference between sessions for the ET group (EMD = 101%, 95% CI [91, 112], $P = 0.9$). There was also a significant interaction between group and time point ($F_{1,\ 432} = 4.9$, $P = 0.03$), with post hoc tests showing that normalized SICF in the UT group was significantly

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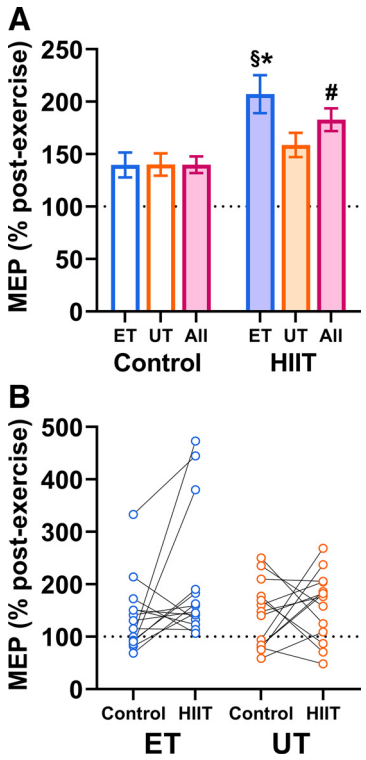

Figure 4. Effect of iTMS on corticospinal excitability following high-intensity interval training (HIIT) or control (no exercise) assessed with single-pulse TMS. Data represent mean test MEP amplitude (*A*) and individual subject means matched between sessions (*B*; ET n = 14, UT n = 14) after iTMS (pooled for 5 and 30 min post) expressed relative to the postexercise time point. Data points above 100% indicate a greater test TMS amplitude, with shaded bars representing a statistically significant difference relative to the postexercise time point. Repeated measures linear mixed model analysis, §$P < 0.001$ compared with control session (ET group); *$P < 0.05$ compared with UT group (HIIT session); #$P < 0.001$ compared with control session (groups combined). ET, endurance-trained; iTMS, I-wave periodicity repetitive TMS; MEP, motor evoked potential; TMS, transcranial magnetic stimulation; UT, untrained.

greater 30 min after iTMS compared with 5 min after iTMS (EMD = 113%, 95% CI [103, 124], P = 0.01) In contrast, there was no difference between time points for the ET group (EMD = 97%, 95% CI [88, 107], P = 0.5). ICF analysis revealed no effect of group ($F_{1,\ 18}$ = 1.0, P = 0.3) or time point ($F_{1,\ 336}$ = 0.02, P = 0.9), but the change in ICF with iTMS for all participants (both groups combined) was greater in the control session compared with the HIIT exercise session (EMD = 84%, 95% CI [76, 94], $F_{1,\ 278}$ = 10, P = 0.002; Fig. 5*C*). No other effects or interactions were present.

Correlations between Corticospinal and M1 Intracortical Excitability after Exercise and iTMS

Spearman's analyses were used to assess correlations between normalized TMS measures (test TMS, SICI, SICF, ICF) recorded after the exercise block and changes in test MEP amplitude with iTMS. For the control session, there was a significant negative correlation between normalized test MEP values recorded after exercise and test MEP values recorded after iTMS (r = −0.60, P = 0.002). On a group level, the same comparison was significant for the ET (r = −0.60, P = 0.03) but not UT (r = −0.50, P = 0.08) group. For the HIIT session, there were no significant correlations between postexercise and post-iTMS measures (all $P > 0.2$). Further correlation analyses were utilized to compare each of the paired-pulse measures recorded after iTMS (normalized to postexercise) to the test MEP recorded after iTMS. The only significant correlation was between normalized measures of SICF and test MEP in the HIIT session, where there was a significant positive correlation in the ET (r = 0.70, P = 0.02) but not the UT (r = −0.3, P = 0.2) group.

Correlations between Muscle Activity during Exercise and TMS Measures

Spearman's analyses revealed no significant correlations between any of the EMG measures of muscle activity (number of bursts, mean burst amplitude, active time and summed area of activity) and TMS measures of excitability recorded after exercise (all $P > 0.2$). Correlation analyses with test MEP amplitude recorded after iTMS (plasticity effect) revealed a significant moderate correlation between number of bursts and normalized MEP amplitude with groups combined (r = −0.41, P = 0.04), as well as between total burst time and normalized MEP amplitude for the UT (r = −0.60, P = 0.03) but not the ET (r = −0.04, P = 0.9) group.

DISCUSSION

This study investigated differences between endurance-trained cyclists and untrained individuals in TMS-induced plasticity following an acute bout of HIIT exercise. The primary finding was that of increased TMS-induced plasticity following HIIT exercise (compared with no-exercise), an effect that was larger in endurance-trained cyclists.

Changes in Corticospinal and M1 Intracortical Excitability with Acute Exercise

Numerous studies have used TMS to examine the change in M1 after an acute bout of exercise. Although the majority of these suggest that corticospinal excitability is unaffected (14–16, 19, 36, 39, 40), several have instead shown increased excitability, but only when exercise was of moderate intensity or greater (40, 41). Furthermore, a recent study by Lulic et al. (2) demonstrated that acute exercise upregulates corticospinal excitability in individuals who are highly active, but not in people with low physical activity levels. In contrast, the present findings demonstrate no effect of acute HIIT exercise on corticospinal excitability regardless of exercise history. A possible reason for this discrepancy between the current study and that of Lulic and colleagues is that each implemented a different intensity of acute exercise. Lulic et al. (2) used continuous moderate-intensity aerobic activity, whereas the present study involved 20 min of HIIT cycling. Although we found no statistically significant change in MEP amplitude with exercise, normalized MEP amplitudes were significantly reduced after HIIT compared with control in endurance-trained cyclists. This may indicate a potential lowering of motor cortical excitability after HIIT exercise, but this is confounded by a larger MEP amplitude in endurance-trained cyclists after the control exercise. The reason for this elevated MEP in the resting control session is

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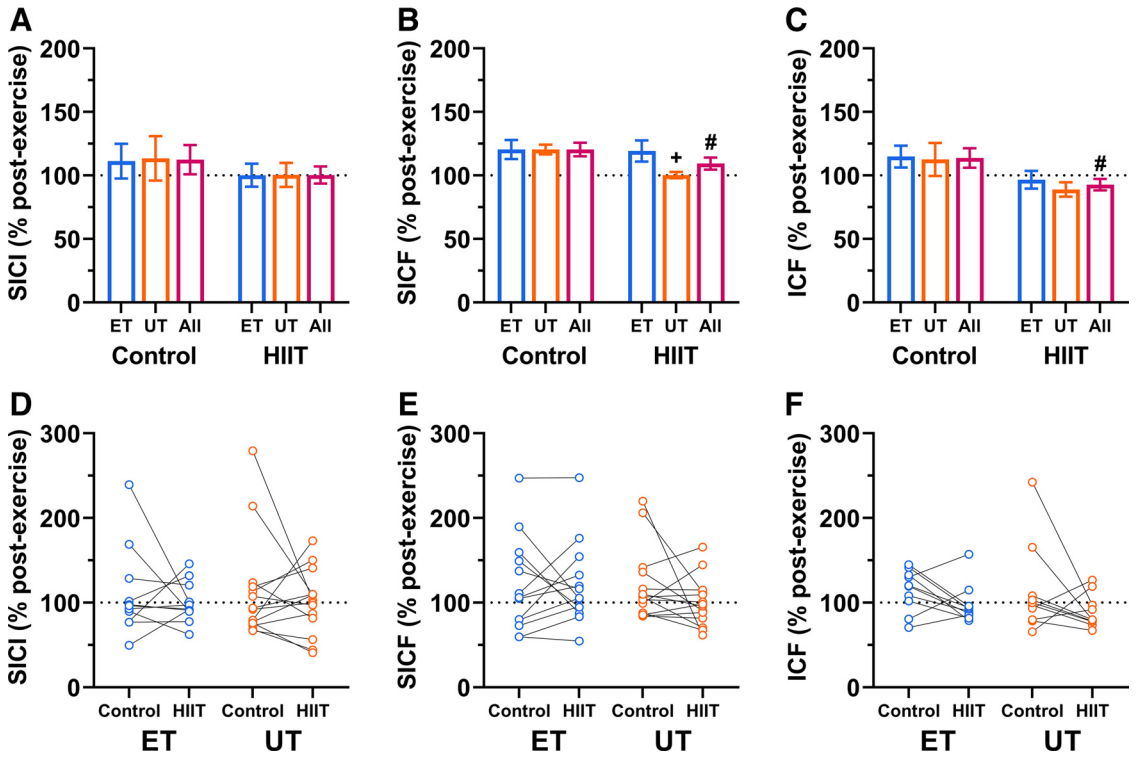

Figure 5. Effect of iTMS on intracortical excitability following high-intensity interval training (HIIT) or control (no exercise) assessed with paired-pulse TMS. Data represent mean response amplitudes (*top row*) and individual subject means matched between sessions (*bottom row*) after iTMS (pooled for 5 and 30 min) for SICI (*A* and *D*; ET $n = 11$, UT $n = 14$), SICF (*B* and *E*; ET $n = 13$, UT $n = 14$), and ICF (*C* and *F*; ET $n = 10$, UT $n = 10$) expressed relative to the postexercise time point. Data points above 100% indicate a reduction of SICI (*A* and *D*) and greater SICF (*B* and *E*), and ICF (*C* and *F*). Repeated measures linear mixed model analyses, #$P = 0.04$ (SICF) and 0.002 (ICF) compared with control session (groups combined); +$P = 0.002$ compared with control session (UT group). ET, endurance-trained; ICF, intracortical facilitation; iTMS, I-wave periodicity repetitive TMS; SICF, short-interval intracortical facilitation; TMS, transcranial magnetic stimulation; UT, untrained.

unclear, but this effect is likely influenced by two endurance-trained subjects who showed ~250% increase in MEP after 20 min of rest.

There is now a considerable number of studies to indicate that acute exercise transiently downregulates $GABA_A$-mediated SICI (19, 33, 42–44). It is therefore surprising that the present results indicate no change in SICI with HIIT in endurance-trained or untrained participants. This outcome is also in contrast to previous findings showing that acute exercise decreases SICI in both high- and low-active individuals (2). Despite this, an insensitivity of SICI to exercise has also been reported following moderate-intensity continuous (36) and HIIT (41) exercise. Interestingly, a recent study by Neva et al. (23) showed exercise-induced disinhibition of SICI, but only when SICI circuits were selectively targeted with anterior-posterior (AP)-induced current, and not when a conventional PA-induced current was used (as in the present study). These findings suggest that examining SICI with a PA current may be less sensitive for detecting changes in SICI circuits with exercise.

In contrast with SICI, relatively few studies have investigated the influence of acute exercise on intracortical facilitation (both SICF and ICF). Current evidence suggests that SICF may be either upregulated (39) or not modulated (2, 44) with low-moderate intensity continuous exercise. Evidence surrounding the modulation of ICF with exercise is similarly variable. For example, Singh and colleagues (42) demonstrated an increase in ICF after exercise in a population of moderately active individuals, whereas Lulic et al. (2) indicated a decrease in ICF with exercise in groups of both high and low levels of habitual physical activity. Nonetheless, our findings in endurance-trained and untrained participants demonstrate that acute HIIT exercise does not modulate M1 intracortical facilitatory circuits.

TMS-Induced Plasticity following HIIT Exercise in Endurance-Trained Cyclists

Growing evidence suggests that physical activity has the potential to modulate TMS-induced plasticity. An increase in plasticity following exercise has been demonstrated with various repetitive TMS paradigms such as PAS (15), iTBS (16), and cTBS (14). In support of this, we provide new evidence that acute exercise enhances plasticity using iTMS, which specifically targets intracortical networks responsible for I-wave generation (21, 22, 45). Given that iTMS has no effect on spinal excitability assessed by cervicomedullary stimulation (45) and is thought to be a form of spike-timing-dependent (Hebbian) plasticity within intracortical networks (21, 22), our findings suggest that acute exercise upregulates the neuroplastic potential of these I-wave generating networks.

In a novel finding and supporting our hypothesis, outcomes of the present study demonstrate that TMS-induced plasticity following acute HIIT is enhanced in endurance-trained cyclists. This finding suggests that highly active

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individuals demonstrate greater LTP-like plasticity following acute exercise. The potential mechanisms that contribute to this effect are unknown, but likely stem from chronic adaptations to the physiological processes that occur with acute exercise. For example, acute exercise is known to enhance levels of lactate, brain derived-neurotrophic factor (BDNF), and uncarboxylated osteocalcin (41, 46, 47), along with increases in neuromodulatory transmitters such as dopamine (for review, see Ref. 11), which are important mediators of LTP/synaptic plasticity (8, 48, 49). In addition, long-term exercise results in increased cerebral blood flow through angiogenesis (5, 6), and the acute exercise-induced upregulation of BDNF is greater in trained individuals (50). Although we are unable to determine the mechanisms of increased plasticity in the current study, our findings suggest that the neurophysiological adaptations resulting from long-term training have a facilitatory benefit to the cortical response to acute exercise, i.e., increased plasticity in endurance-trained cyclists after acute exercise.

Factors That Contribute to Increased TMS-Induced Plasticity with Exercise

Apart from adaptations due to long-term endurance exercise, a number of other neurophysiological factors could potentially contribute to an increase in TMS-induced plasticity after acute exercise in endurance-trained cyclists. For example, there could be neurophysiological differences between endurance-trained and untrained participants at baseline. However, participant groups were well matched for baseline TMS data, with no difference between groups for any neurophysiological measure. Furthermore, there could be neurophysiological differences between endurance-trained and untrained participants with exercise. This was assessed in three ways. First, we examined the change in single and paired-pulse TMS after exercise, but there were no differences in the change in any TMS response with HIIT exercise between participant groups. Second, we quantified the magnitude of hand muscle activity during exercise, as prior activity may contribute to differences in the effects of iTMS postexercise. Although there was greater muscle activity (number of bursts, active time and summed area of activity) during the exercise compared with the control session (no exercise), there was no difference in muscle activity during exercise between endurance-trained and untrained participants (Table 2), suggesting that this does not contribute to the increased TMS-induced plasticity with exercise in endurance-trained cyclists. Third, we compared the physiological (power to weight ratio, exercise intensity) and psychological (RPE, affect score) response between groups during exercise. However, there were no differences in the heart rate response or perceived exertion between groups, although there was a greater affect score in endurance-trained cyclists, indicating that this group maintained a more positive emotional expression during exercise (35). Although speculative, the possibility exists that a more positive emotional state during exercise in the endurance-trained cyclists had implications for dopaminergic transmission, thereby stimulating plasticity (11, 51, 52). We cannot exclude the possibility that our findings may have differed if an alternative acute exercise intervention (e.g., treadmill running) was utilized. Furthermore, for logistical reasons, all participants were aware of which session they would be completing before entering the laboratory, and it is possible that this knowledge may have influenced the response to exercise, especially if the expectation of exercise provoked stressful or positive emotional states. Alternatively, there could be other circadian (chronotype), attentional (focused/unfocussed), pharmacological (medications) or genetic (e.g., BDNF polymorphisms) factors that might influence plasticity in different participant populations (for review, see Ref. 53). Finally, it is important to mention that this study did not utilize a neuronavigation system, which would have provided a more robust method of ensuring consistent coil position and orientation. However, the influence of these factors is unlikely to influence one group more than the other. Nonetheless, their potential contribution to M1 plasticity after exercise should be explored in future studies.

In conclusion, this study is the first to demonstrate that TMS-induced M1 plasticity is greater in endurance-trained cyclists after an acute bout of exercise. Further, we support previous findings that TMS-induced plasticity is greater following acute HIIT exercise. These findings add to the extensive body of literature surrounding the positive effect of exercise on neuroplasticity and indicate that exercise history is an important consideration for understanding neuroplasticity.

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the authors.

AUTHOR CONTRIBUTIONS

B.J.H., G.M.O., and J.G.S. conceived and designed research; B.J.H. performed experiments; B.J.H. analyzed data; B.J.H., G.M.O., and J.G.S. interpreted results of experiments; B.J.H. prepared figures; B.J.H., G.M.O., and J.G.S. drafted manuscript; B.J.H., G.M.O., S.K.S., and J.G.S. edited and revised manuscript; B.J.H., G.M.O., S.K.S., and J.G.S. approved final version of manuscript.

REFERENCES

1. **Hassanlouei H**, **Sundberg CW**, **Smith AE**, **Kuplic A**, **Hunter SK.** Physical activity modulates corticospinal excitability of the lower limb in young and old adults. *J Appl Physiol (1985)* 123: 364–374, 2017. doi:10.1152/japplphysiol.01078.2016.
2. **Lulic T**, **El-Sayes J**, **Fassett H**, **Nelson A.** Physical activity levels determine exercise-induced changes in brain excitability. *PLOS One* 12: e0173672, 2017. doi:10.1371/journal.pone.0173672.
3. **Cirillo J**, **Lavender AP**, **Ridding MC**, **Semmler JG.** Motor cortex plasticity induced by paired associative stimulation is enhanced in physically active individuals. *J Physiol* 587: 5831–5842, 2009. doi:10.1113/jphysiol.2009.181834.
4. **Sanes JN**, **Donoghue JP.** Plasticity and primary motor cortex. *Annu Rev Neurosci* 23: 393–415, 2000. doi:10.1146/annurev.neuro.23.1.393.
5. **Kleim JA**, **Cooper NR**, **VandenBerg PM.** Exercise induces angiogenesis but does not alter movement representations within rat motor cortex. *Brain Res* 934: 1–6, 2002. doi:10.1016/s0006-8993(02)02239-4.

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6. **Xiong J**, **Ma L**, **Wang B**, **Narayana S**, **Duff EP**, **Egan GF**, **Fox PT.** Long-term motor training induced changes in regional cerebral blood flow in both task and resting states. *NeuroImage* 45: 75–82, 2009. doi:10.1016/j.neuroimage.2008.11.016.
7. **Currie J**, **Ramsbottom R**, **Ludlow H**, **Nevill A**, **Gilder M.** Cardio-respiratory fitness, habitual physical activity and serum brain derived neurotrophic factor (BDNF) in men and women. *Neurosci Lett* 451: 152–155, 2009. doi:10.1016/j.neulet.2008.12.043.
8. **Schinder AF**, **Poo M-M.** The neurotrophin hypothesis for synaptic plasticity. *Trends Neurosci* 23: 639–645, 2000. doi:10.1016/S0166-2236(00)01672-6.
9. **Sibley BA**, **Etnier JL.** The effects of physical activity on cognition in children: a meta-analysis. *Med Sci Sports Exerc* 34: S214, 2002. doi:10.1097/00005768-200205001-01198.
10. **McDonnell MN**, **Smith AE**, **Mackintosh SF.** Aerobic exercise to improve cognitive function in adults with neurological disorders: a systematic review. *Arch Phys Med Rehabil* 92: 1044–1052, 2011. doi:10.1016/j.apmr.2011.01.021.
11. **Taubert M**, **Villringer A**, **Lehmann N.** Endurance exercise as an "endogenous" neuro-enhancement strategy to facilitate motor learning. *Front Hum Neurosci* 9: 692, 2015. doi:10.3389/fnhum.2015.00692.
12. **Roig M**, **Skriver K**, **Lundbye-Jensen J**, **Kiens B**, **Nielsen JB.** A single bout of exercise improves motor memory. *PLOS One* 7: e44594, 2012. doi:10.1371/journal.pone.0044594.
13. **Baltar A**, **Nogueira F**, **Marques D**, **Carneiro M**, **Monte-Silva K.** Evidence of the homeostatic regulation with the combination of transcranial direct current stimulation and physical activity. *Am J Phys Med Rehabil* 97: 727–733, 2018. doi:10.1097/phm.0000000000000956.
14. **McDonnell MN**, **Buckley JD**, **Opie GM**, **Ridding MC**, **Semmler JG.** A single bout of aerobic exercise promotes motor cortical neuroplasticity. *J Appl Physiol (1985)* 114: 1174–1182, 2013. doi:10.1152/japplphysiol.01378.2012.
15. **Mang CS**, **Snow NJ**, **Campbell KL**, **Ross CJ**, **Boyd LA.** A single bout of high-intensity aerobic exercise facilitates response to paired associative stimulation and promotes sequence-specific implicit motor learning. *J Appl Physiol (1985)* 117: 1325–1336, 2014. doi:10.1152/japplphysiol.00498.2014.
16. **Andrews SC**, **Curtin D**, **Hawi Z**, **Wongtrakun J**, **Stout JC**, **Coxon JP.** Intensity matters: high-intensity interval exercise enhances motor cortex plasticity more than moderate exercise. *Cereb Cortex* 30: 101–112, 2020. doi:10.1093/cercor/bhz075.
17. **Mellow ML**, **Goldsworthy MR**, **Coussens S**, **Smith AE.** Acute aerobic exercise and neuroplasticity of the motor cortex: a systematic review. *J Sci Med Sport* 23: 408–414, 2020. doi:10.1016/j.jsams.2019.10.015.
18. **Rozand V**, **Senefeld JW**, **Sundberg CW**, **Smith AE**, **Hunter SK.** Differential effects of aging and physical activity on corticospinal excitability of upper and lower limb muscles. *J Neurophysiol* 122: 241–250, 2019. doi:10.1152/jn.00077.2019.
19. **Smith AE**, **Goldsworthy MR**, **Garside T**, **Wood FM**, **Ridding MC.** The influence of a single bout of aerobic exercise on short-interval intracortical excitability. *Exp Brain Res* 232: 1875–1882, 2014. doi:10.1007/s00221-014-3879-z.
20. **Singh AM**, **Neva JL**, **Staines WR.** Acute exercise enhances the response to paired associative stimulation-induced plasticity in the primary motor cortex. *Exp Brain Res* 232: 3675–3685, 2014. doi:10.1007/s00221-014-4049-z.
21. **Thickbroom GW**, **Byrnes ML**, **Edwards DJ**, **Mastaglia FL.** Repetitive paired-pulse TMS at I-wave periodicity markedly increases corticospinal excitability: a new technique for modulating synaptic plasticity. *Clin Neurophysiol* 117: 61–66, 2006. doi:10.1016/j.clinph.2005.09.010.
22. **Kidgell DJ**, **Mason J**, **Frazer A**, **Pearce AJ.** I-wave periodicity transcranial magnetic stimulation (iTMS) on corticospinal excitability. A systematic review of the literature. *Neuroscience* 322: 262–272, 2016. doi:10.1016/j.neuroscience.2016.02.041.
23. **Neva JL**, **Brown KE**, **Peters S**, **Feldman SJ**, **Mahendran N**, **Boisgontier MP**, **Boyd LA.** Acute exercise modulates the excitability of specific interneurons in human motor cortex. *Neuroscience* 475: 103–116, 2021. doi:10.1016/j.neuroscience.2021.08.032.
24. **Craig CL**, **Marshall AL**, **Sjöström M**, **Bauman AE**, **Booth ML**, **Ainsworth BE**, **Pratt M**, **Ekelund U**, **Yngve A**, **Sallis JF**, **Oja P.** International physical activity questionnaire: 12-country reliability and validity. *Med Sci Sports Exerc* 35: 1381–1395, 2003. doi:10.1249/01.Mss.0000078924.61453.Fb.
25. **Cash RF**, **Benwell NM**, **Murray K**, **Mastaglia FL**, **Thickbroom GW.** Neuromodulation by paired-pulse TMS at an I-wave interval facilitates multiple I-waves. *Exp Brain Res* 193: 1–7, 2009. doi:10.1007/s00221-008-1590-7.
26. **Thomas S**, **Reading J**, **Shephard RJ.** Revision of the physical activity readiness questionnaire (PAR-Q). *Can J Sport Sci* 17: 338–345, 1992.
27. **Sale MV**, **Ridding MC**, **Nordstrom MA.** Cortisol inhibits neuroplasticity induction in human motor cortex. *J Neurosci* 28: 8285–8293, 2008. doi:10.1523/JNEUROSCI.1963-08.2008.
28. **Hermens HJ**, **Freriks B**, **Disselhorst-Klug C**, **Rau G.** Development of recommendations for SEMG sensors and sensor placement procedures. *J Electromyogr Kinesiol* 10: 361–374, 2000. doi:10.1016/s1050-6411(00)00027-4.
29. **Kujirai T**, **Caramia MD**, **Rothwell JC**, **Day BL**, **Thompson PD**, **Ferbert A**, **Wroe S**, **Asselman P**, **Marsden CD.** Corticocortical inhibition in human motor cortex. *J Physiol* 471: 501–519, 1993. doi:10.1113/jphysiol.1993.sp019912.
30. **Opie GM**, **Semmler JG.** Modulation of short- and long-interval intracortical inhibition with increasing motor evoked potential amplitude in a human hand muscle. *Clin Neurophysiol* 125: 1440–1450, 2014. doi:10.1016/j.clinph.2013.11.015.
31. **Ziemann U**, **Tergau F**, **Wassermann EM**, **Wischer S**, **Hildebrandt J**, **Paulus W.** Demonstration of facilitatory I wave interaction in the human motor cortex by paired transcranial magnetic stimulation. *J Physiol* 511: 181–190, 1998. doi:10.1111/j.1469-7793.1998.181bi.x.
32. **Wagle-Shukla A**, **Ni Z**, **Gunraj CA**, **Bahl N**, **Chen R.** Effects of short interval intracortical inhibition and intracortical facilitation on short interval intracortical facilitation in human primary motor cortex. *J Physiol* 587: 5665–5678, 2009. doi:10.1113/jphysiol.2009.181446.
33. **Stavrinos EL**, **Coxon JP.** High-intensity interval exercise promotes motor cortex disinhibition and early motor skill consolidation. *J Cogn Neurosci* 29: 593–604, 2017. doi:10.1162/jocn_a_01078.
34. **Borg G.** Perceived exertion as an indicator of somatic stress. *Scand J Rehabil Med* 2: 92–98, 1970.
35. **Hardy CJ**, **Rejeski WJ.** Not what, but how one feels: the measurement of affect during exercise. *J Sport Exerc Psychol* 11: 304–317, 1989. doi:10.1123/jsep.11.3.304.
36. **Mooney RA**, **Coxon JP**, **Cirillo J**, **Glenny H**, **Gant N**, **Byblow WD.** Acute aerobic exercise modulates primary motor cortex inhibition. *Exp Brain Res* 234: 3669–3676, 2016. doi:10.1007/s00221-016-4767-5.
37. **Mukaka MM.** Statistics corner: a guide to appropriate use of correlation coefficient in medical research. *Malawi Med J* 24: 69–71, 2012.
38. **Opie GM**, **Pourmajidian M**, **Ziemann U**, **Semmler JG.** Investigating the influence of paired-associative stimulation on multi-session skill acquisition and retention in older adults. *Clin Neurophysiol* 131: 1497–1507, 2020. doi:10.1016/j.clinph.2020.04.010.
39. **Neva JL**, **Brown KE**, **Mang CS**, **Francisco BA**, **Boyd LA.** An acute bout of exercise modulates both intracortical and interhemispheric excitability. *Eur J Neurosci* 45: 1343–1355, 2017. doi:10.1111/ejn.13569.
40. **Opie GM**, **Semmler JG.** Acute exercise at different intensities influences corticomotor excitability and performance of a ballistic thumb training task. *Neuroscience* 412: 29–39, 2019. doi:10.1016/j.neuroscience.2019.05.049.
41. **Nicolini C**, **Michalski B**, **Toepp SL**, **Turco CV**, **D'Hoine T**, **Harasym D**, **Gibala MJ**, **Fahnestock M**, **Nelson AJ.** A single bout of high-intensity interval exercise increases corticospinal excitability, brain-derived neurotrophic factor, and uncarboxylated osteolcalcin in sedentary, healthy males. *Neuroscience* 437: 242–255, 2020. doi:10.1016/j.neuroscience.2020.03.042.
42. **Singh AM**, **Duncan RE**, **Neva JL**, **Staines WR.** Aerobic exercise modulates intracortical inhibition and facilitation in a nonexercised upper limb muscle. *BMC Sports Sci Med Rehabil* 6: 23, 2014. doi:10.1186/2052-1847-6-23.
43. **Yamaguchi T**, **Fujiwara T**, **Liu W**, **Liu M.** Effects of pedaling exercise on the intracortical inhibition of cortical leg area. *Exp Brain Res* 218: 401–406, 2012. doi:10.1007/s00221-012-3026-7.
44. **Yamazaki Y**, **Sato D**, **Yamashiro K**, **Nakano S**, **Onishi H**, **Maruyama A.** Acute low-intensity aerobic exercise modulates intracortical inhibitory and excitatory circuits in an exercised and a non-exercised muscle in the primary motor cortex. *Front Physiol* 10: 1361, 2019. doi:10.3389/fphys.2019.01361.
45. **Di Lazzaro V**, **Thickbroom GW**, **Pilato F**, **Profice P**, **Dileone M**, **Mazzone P**, **Insola A**, **Ranieri F**, **Tonali PA**, **Rothwell JC.** Direct

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demonstration of the effects of repetitive paired-pulse transcranial magnetic stimulation at I-wave periodicity. *Clin Neurophysiol* 118: 1193–1197, 2007. doi:10.1016/j.clinph.2007.02.020.

46. **Ferris LT**, **Williams JS**, **Shen CL.** The effect of acute exercise on serum brain-derived neurotrophic factor levels and cognitive function. *Med Sci Sports Exerc* 39: 728–734, 2007. doi:10.1249/mss.0b013e31802f04c7.
47. **Etnier JL**, **Wideman L**, **Labban JD**, **Piepmeier AT**, **Pendleton DM**, **Dvorak KK**, **Becofsky K.** The effects of acute exercise on memory and brain-derived neurotrophic factor (BDNF). *J Sport Exerc Psychol* 38: 331–340, 2016. doi:10.1123/jsep.2015-0335.
48. **Oury F**, **Khrimian L**, **Denny CA**, **Gardin A**, **Chamouni A**, **Goeden N**, **Huang YY**, **Lee H**, **Srinivas P**, **Gao XB**, **Suyama S**, **Langer T**, **Mann JJ**, **Horvath TL**, **Bonnin A**, **Karsenty G.** Maternal and offspring pools of osteocalcin influence brain development and functions. *Cell* 155: 228–241, 2013. doi:10.1016/j.cell.2013.08.042.
49. **Kuipers SD**, **Bramham CR.** Brain-derived neurotrophic factor mechanisms and function in adult synaptic plasticity: new insights and implications for therapy. *Curr Opin Drug Discov Devel* 9: 580–586, 2006.
50. **Zoladz JA**, **Pilc A**, **Majerczak J**, **Grandys M**, **Zapart-Bukowska J**, **Duda K.** Endurance training increases plasma brain-derived neurotrophic factor concentration in young healthy men. *J Physiol Pharmacol* 59 *Suppl* 7: 119–132, 2008.
51. **Lin TW**, **Kuo YM.** Exercise benefits brain function: the monoamine connection. *Brain Sci* 3: 39–53, 2013. doi:10.3390/brainsci3010039.
52. **Kuo M-F**, **Paulus W**, **Nitsche MA.** Boosting focally-induced brain plasticity by dopamine. *Cereb Cortex* 18: 648–651, 2008. doi:10.1093/cercor/bhm098.
53. **Ridding MC**, **Ziemann U.** Determinants of the induction of cortical plasticity by non-invasive brain stimulation in healthy subjects. *J Physiol* 588: 2291–2304, 2010. doi:10.1113/jphysiol.2010.190314.

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