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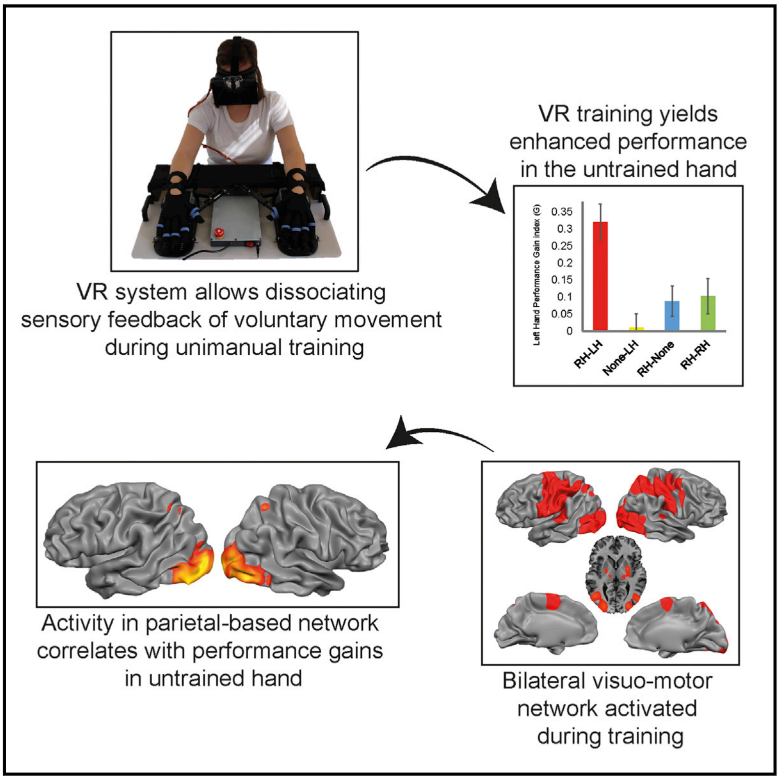
Neural Network Underlying Intermanual Skill Transfer in Humans

Graphical Abstract



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In Brief

Physical practice is important when learning a new motor skill. Ossmy and Mukamel demonstrate a training scheme in the absence of voluntary physical training and establish a link between neural activity during training and subsequent learning. Their results may have practical implications for rehabilitation of patients with upper-extremity hemiparesis.

Highlights

- Unimanual training also enhances performance in the untrained hand (cross-education)
- Real-time manipulation of visual feedback enhances magnitude of cross-education
- Yoking movement of untrained to trained hand further increases cross-education
- Functional connectivity with SPL during training predicts cross-education

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Neural Network Underlying Intermanual Skill Transfer in Humans

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SUMMARY

Physical practice with one hand results in performance gains of the other (un-practiced) hand, yet the role of sensory feedback and underlying neurophysiology is unclear. Healthy subjects learned sequences of finger movements by physical training with their right hand while receiving real-time movement-based visual feedback via 3D virtual reality devices as if their immobile left hand was training. This manipulation resulted in significantly enhanced performance gain with the immobile hand, which was further increased when left-hand fingers were yoked to passively follow right-hand voluntary movements. Neuroimaging data show that, during training with manipulated visual feedback, activity in the left and right superior parietal lobule and their degree of coupling with motor and visual cortex, respectively, correlate with subsequent left-hand performance gain. These results point to a neural network subserving short-term motor skill learning and may have implications for developing new approaches for learning and rehabilitation in patients with unilateral motor deficits.

INTRODUCTION

It is common wisdom that “practice makes perfect”; however, what constitutes an optimal practice regime when learning a new skill is not clear. In the domain of motor skills, for example, when learning to dribble a basketball, physical training with the relevant effector obviously plays a crucial role. Nonetheless, research over the past decades has recognized that sensory feedback and mental imagery play a significant role in the learning process (Nyberg et al., 2006; Sigrist et al., 2013; Wolpert et al., 2011). In the case of vision, it has been shown that even in the absence of physical training, mere observation of someone else performing a motor task is sufficient to introduce significant gains in subsequent performance of the observer (Bird et al., 2005; Cross et al., 2009; Kelly et al., 2003; Mattar and Gribble, 2005; Nojima et al., 2015; Vogt and Thomaschke, 2007; Ossmy and Mukamel, 2016). Furthermore, passive limb movement has also been shown to facilitate learning (Beets et al., 2012; Darainy et al., 2013; Vahdat et al., 2014; Wong et al., 2012). Finally, physical training with one hand is known to result in significant performance gains in the opposite (untrained) hand—a phenomenon termed intermanual transfer or cross-education (Ruddy and Carson, 2013). Intermanual transfer has been reported as early as 1894, showing that unilateral strength training of a single limb increases the strength of the contralateral (untrained) homologous muscle group (Scripture et al., 1894). Since then, this effect has been demonstrated across multiple motor tasks (Anguera et al., 2007; Brass et al., 2001; Camus et al., 2009; Carroll et al., 2006; Criscimagna-Hemminger et al., 2003; Farthing et al., 2007; Lee et al., 2010; Malfait and Ostry, 2004; Perez and Cohen, 2008; Perez et al., 2007; Sainburg and Wang, 2002) and is suggested to occur through plastic changes in the brain that are not confined to the specific neural networks controlling the physically trained effector (e.g., plastic changes also in motor cortex ipsilateral to the active hand [Duque et al., 2008; Hortobágyi et al., 2003; Muellbacher et al., 2000; Obayashi, 2004]). Enhancing the behavioral effect of intermanual transfer and elucidating its underlying neural mechanism has important implications for rehabilitation of patients with unimanual deficits (Hendy et al., 2012; Ramachandran and Altschuler, 2009) in which direct training of the affected hand is difficult.

Given that visual input, physical training, and passive movement play a significant role in performance and intermanual transfer of motor skills, research in recent years examined the behavioral and neural consequences of training with manipulated visual feedback (Halsband and Lange, 2006). In particular, unimanual training with mirrored visual feedback (as if the opposite, passive hand, is training) has been shown to enhance transfer to the opposite hand and increase excitability of primary motor cortex (M1) ipsilateral to the physically trained hand (Garry et al., 2005; Hamzei et al., 2012; Nojima et al., 2012). Nonetheless, much less is known at the whole-brain network level and how inter-regional coupling during such training correlates with subsequent behavioral changes in performance. Additionally, at the behavioral level, the interaction between manipulated visual feedback and passive movement during training is unknown.

In the present study, we examined intermanual transfer using a novel setup employing 3D virtual reality (VR) devices to control visual feedback of finger movements during unimanual training of healthy adults (experiment 1). By using a novel device, we also examined whether the addition of passive finger movement of the non-physically training hand further enhances the

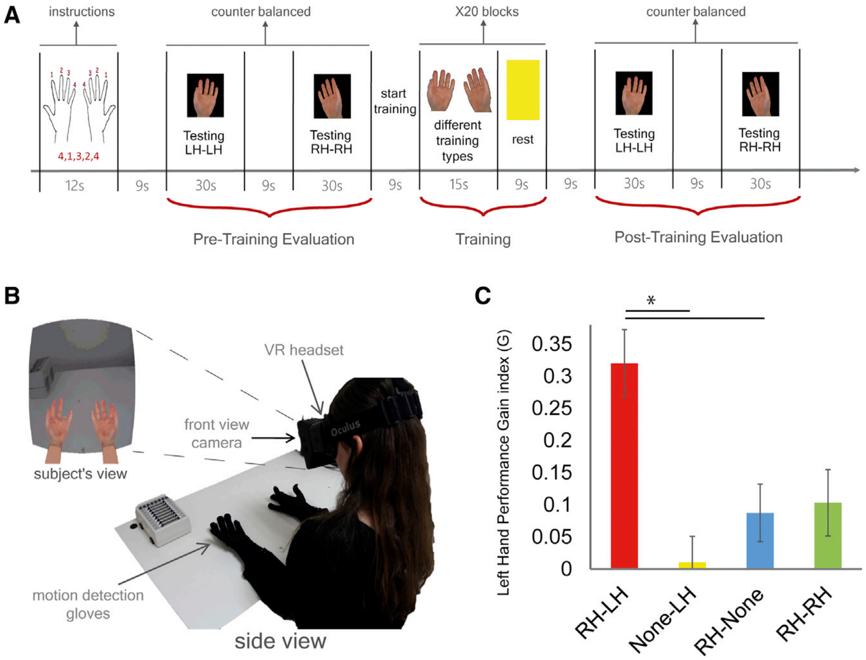

Figure 1. Experiment 1

(A) Schematic illustration of one experimental condition. A unique sequence of five digits was presented together with a sketch of the mapped fingers (instructions). Subjects performed the sequence as accurately and rapidly as possible using their right hand (RH) and their left hand (LH) separately for initial evaluation of performance. Next, subjects were trained under a specific training type and finally repeated the evaluation test again.

(B) Subjects wore a headset and motion sensitive gloves and received visual feedback of virtual hands. The VR devices allowed visual manipulation of online visual feedback. A camera mounted on the headset allowed embedding the virtual hands and subject's view inside a natural environment.

(C) Experiment 1 results. Physical training with the right hand while receiving online visual feedback as if the left hand is moving (RH-LH) resulted in highest left-hand performance gains relative to all other training conditions. Error bars indicate SEM across subjects. For condition acronyms, see Table 1.

intermanual transfer effect (experiment 2). Finally, we used whole-brain functional magnetic resonance imaging (fMRI) to probe the relevant brain regions engaged during such training and examined their degree of inter-regional coupling with respect to subsequent behavioral changes in performance of individual subjects (experiment 3).

RESULTS

A total of 53 subjects participated in one of three different experiments in which they trained to execute rapid sequences of finger movements. Throughout the entire study, subjects could not see their real hands and visual feedback of two virtual hands was provided by the VR setup. Each experimental condition comprised a pre-training performance evaluation stage, a training stage, and a post-training performance evaluation stage. In the evaluation stages, subjects were instructed to perform a unimanual five-digit movement sequence repeatedly as accurately and rapidly as possible (Karni et al., 1998). Performance level was calculated for each hand separately as the number of times within a fixed time window that the subject performed a complete five-digit sequence with no errors. In the training stage, subjects were instructed to perform the same sequence of finger movements at self-pace while sensory feedback was manipulated according to the different experimental conditions (detailed below). An index of performance gains (G) was calculated based on the difference in performance level between the post-training and pre-training evaluation stages for each subject and training condition (see Experimental Procedures).

Visual Manipulation

We used specialized VR devices, including motion sensitive gloves, to detect subject's real hand movements and translate them by customized software to movement of virtual hands presented on the screen. This setup allowed us to decouple the online visual feedback of virtual-hand movements from the subject's real hand movements during the training stage. In each experimental condition, the subject trained on a specific sequence of finger movements (see Figure 1). Training manner varied across four different experimental conditions (see Table 1 and Experimental Procedures): (1) congruent visual feedback—subjects physically trained with their right hand while receiving congruent online visual feedback of right-virtual-hand movement (condition RH-RH); (2) incongruent visual feedback—subjects physically trained with their right hand while receiving online corresponding visual feedback of left-virtual-hand movement (condition RH-LH; see Movie S1); (3) no visual feedback—subjects physically trained with their right hand while no visual feedback was provided (condition RH-None); (4) observation only—subjects passively observed the virtual left hand performing the sequence, while both their real hands were immobile (condition None-LH).

The average number of self-paced full-sequence movements performed by the subjects during training was 106.4 ± 3.8 (mean ± SEM across subjects and training types) and was not significantly different across the different training types (repeated-measures ANOVA [rmANOVA], minimal $p = 0.53$ across subjects). The lack of real hand movement (i.e., left-hand movement in conditions 1–3, and both hands in condition 4) was verified with the motion detection gloves (see Experimental Procedures). This suggests that differences in performance gains across experimental conditions is unlikely due to differences in the amount of movements across training conditions or subliminal hand movements.

Physical training with the right hand (conditions 1–3) yielded significant performance gains in the left (non-practicing) hand

Table 1. Training Conditions Used across the Three Experiments

	Real-Hand Movement		Visual Feedback	
Training Type	Left Hand	Right Hand	Left Virtual Hand	Right Virtual Hand
RH-RH	no movement	active sequence generation	no movement	yoked to real right hand
RH-LH	no movement	active sequence generation	yoked to right-hand movement	no movement
RH-none	no movement	active sequence generation	no virtual hand	no virtual hand
None-RH	no movement	no movement	no movement	simulating sequence execution
None-LH	no movement	no movement	simulating sequence execution	no movement
RH-RH-PM	yoked to right hand	active sequence generation	yoked to left-hand passive movement	yoked to real right hand
RH-LH-PM	yoked to right hand	active sequence generation	yoked to real right hand	no movement
RH-none-PM	yoked to right hand	active sequence generation	no movement	no movement

Acronym notation corresponds to (physical training)-(visual feedback)-(passive movement). RH, right hand; LH, left hand; PM, passive left-hand movement by the device (experiment 2). Thus, condition RH-RH corresponds to training condition in which subjects physically trained with their right hand and received online visual feedback of right-virtual-hand movement. RH-none-PM corresponds with right-hand physical training, without visual feedback, and with left-hand passive movement.

while passive training by left-hand observation (condition 4) yielded performance gains that were not significantly different than zero ($p = 0.7$; unequal variance t test; see Figure 1C and Table S1). Interestingly, we found that training in the incongruent visual feedback condition (RH-LH) resulted in the strongest intermanual transfer. Left-hand performance gain in this condition was significantly higher than gains obtained following training with congruent (RH-RH) visual feedback ($p < 0.05$; two tailed paired t test) and training without visual feedback (RH-None; $p < 0.05$; two-tailed paired t test; see Figure 1C). Additionally, performance gain in the incongruent visual feedback condition (RH-LH) was higher than the sum of performance gains obtained during right-hand training without visual feedback (RH-None) and training by left-hand observation (None-LH) conditions separately ($p < 0.01$). This implies that performance gains in the left hand are non-linearly enhanced when right-hand training is supplemented with left-hand visual feedback that is controlled by the subject.

Passive Movement

In experiment 1, we demonstrated that intermanual transfer of a motor skill can be enhanced by providing subject-controlled incongruent visual feedback during training. In experiment 2, we examined whether passive left-hand movement can further enhance this transfer effect. To this end, throughout all training periods, subjects' hands were placed inside a custom-built device that controls left-hand finger movement (Figure 2A and Experimental Procedures). The device allows subjects to freely perform right-hand finger movements (flexion/extension), while corresponding left-hand finger movement is yoked to right-hand finger movements (Movie S2). Voluntary left-hand movement is impossible. Thus, for example, voluntary right-hand index finger flexion/extension results in an immediate passive flexion/extension of the left-hand index finger by the device. A new set of subjects ($n = 18$) completed five experimental training conditions: RH-LH and RH-None (similar to experiment 1) and another three experimental conditions (RH-RH-PM, RH-LH-PM, and RH-None-PM), in which right-hand active finger movement resulted in yoked passive movement of the corresponding left-hand finger through the device. Thus, condition RH-LH-PM corresponds to right-hand physical training with incongruent visual feedback (similar to condition RH-LH from experiment 1) with the addition of passive movement (PM) of the left hand. In this setup, subjects performed 89.7 ± 8.6 full-sequence movements on average during training (mean across subjects and training types). Number of movements performed during training was not significantly different across the different training types (rmANOVA; minimal $p = 0.61$ across subjects). The average number of movements performed in this setup is smaller than that of experiment 1 probably due to the use of the device which made subject movements slower (Table S2). Importantly, since our performance gain index (G) depends also on total number of movements performed, G index comparisons were only performed across conditions within a given experiment and not across experiments.

Our results from experiment 1 demonstrate that, during right-hand physical training, the addition of congruent right-virtual-hand visual feedback did not significantly improve left-hand performance gains (experiment 1, RH-RH versus RH-None). In the current experiment, we find that in the absence of visual feedback the addition of left-hand passive movement does not significantly improve left-hand performance gains either (RH-None-PM versus RH-None; $p = 0.41$; see Figure 2B and Table S2). Furthermore, we found that the addition of both congruent right-virtual-hand visual feedback and passive left-hand movement together do not introduce significantly different gains in left-hand performance (RH-RH-PM versus RH-None; $p = 0.38$).

However, this was not true in the case of incongruent visual feedback. The addition of incongruent visual feedback during right-hand training significantly improved left-hand performance gains (RH-LH > RH-None; $p = 3 \times 10^{-4}$), thus replicating the results from experiment 1. As mentioned above, the addition of passive movement (either with or without congruent visual feedback) did not result in significant left-hand performance gains. Importantly, the addition of passive movement to the incongruent condition (RH-LH-PM) led to the highest left-hand performance gains (see Figure 2B), which was significantly higher compared to all other training types ($F(4,85) = 6.23$, $p < 0.001$; rmANOVA). Performance gain obtained in the RH-LH-PM training condition was even higher than that obtained in the

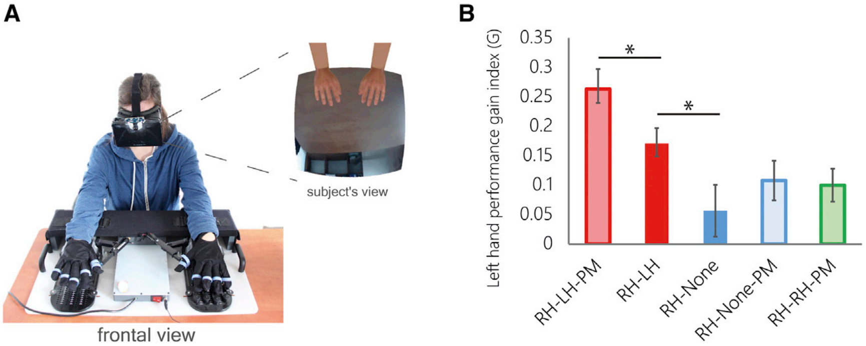

Figure 2. Experiment 2

(A) The custom-built device was added to the virtual reality setup in order to produce passive left-hand finger movement that is yoked to right-hand voluntary finger movement during training. The online manipulation of virtual hands remained similar to experiment 1 (see Figure 1).

(B) The highest left-hand performance gain was obtained when incongruent visual feedback was combined with passive left-hand movement (RH-LH-PM). This improvement was significantly higher than condition RH-LH, which was the most effective training type in experiment 1. Error bars indicate SEM across subjects.

RH-LH training condition (RH-LH-PM > RH-LH, $p = 0.003$). As mentioned above, and similar to experiment 1, such differences in performance gains across training conditions cannot be explained by differences in the number of self-paced movements or subliminal movement of the untrained hand (see Experimental Procedures).

Neuroimaging: Individual Region Analysis

We explored putative brain regions underlying the enhanced intermanual transfer effects we obtained in the previous behavioral studies. To this end, we acquired whole-brain fMRI data from an additional set of subjects ($n = 18$) who completed five different training conditions (similar to experiment 1; see Figure 1A). Since the passive-hand movement device used in experiment 2 is not MR compatible, it could not be used in the current experiment. Subjects underwent the following experimental conditions: RH-RH, RH-LH, RH-None, None-LH, and None-RH (the last two conditions are training by observation only, in which subjects passively observed the virtual left or right hand perform the sequence, respectively; see Ossmy and Mukamel, 2016 for further details). Subjects performed a total number of 126 ± 11.6 full-sequence movements during each training type (averaged across subjects and training types), which was not significantly different across training conditions (minimal $p = 0.34$ across subjects; rmANOVA). Similar to experiment 1, training condition RH-LH was significantly more effective than all other training conditions with respect to transfer of performance gains to the untrained left hand ($p < 0.05$; rmANOVA) (see Figure 3A and Table S3). Furthermore, the combination of right-hand physical training with left-hand observation (RH-LH) was better than each training type separately (RH-None, and None-LH) and better than the sum of both of them ($p = 0.03$; one-tailed paired t test). Conversely, the combination of right-hand physical training with right-hand observation (RH-RH) did not yield significantly different left-hand performance gains than each training type separately (RH-None, and None-RH; $F(2,51) = 0.25$, $p = 0.77$; rmANOVA). This suggests that the congruency of visual feedback during physical training with the right hand plays an important role in intermanual transfer.

At the neural level, we first detected relevant brain regions engaged during training with incongruent feedback. To this end, we performed a general linear model (GLM) analysis on the fMRI data obtained during the training stage by using the contrast: RH-LH > RH-None (see Figure 3B). This analysis revealed a network of four regions including right superior parietal lobule (R-SPL), left superior parietal lobule (L-SPL), and bilateral occipito-temporal visual regions (R-Visual and L-Visual). Activity in these regions during training was then examined with respect to corresponding behavioral changes in left-hand performance (see Experimental Procedures). We found that, of the four regions of interest (ROIs), only activity in left and right SPL correlated with the performance gain value across individual subjects (see Figure 3C; $r = 0.68$ and $r = 0.62$, respectively; $p < 0.05$ corrected for number of ROIs). Activity, in the right or left visual regions was not significant ($r = 0.41$, $p = 0.09$ and $r = 0.37$, $p = 0.13$, respectively). These results demonstrate that fMRI activity level in both SPL during training with incongruent visual feedback is a good predictor for subsequent left-hand performance gains across individual subjects.

We also performed a similar analysis using data from the congruent training condition (RH-RH > RH-None; Figure 4A and Experimental Procedures). This analysis revealed three ROIs (left and right visual cortex, and right SPL); however, the correlation between the fMRI activity level in these regions during RH-RH training with the corresponding left-hand performance gains was not significant (uncorrected minimal $p = 0.12$ across regions was in R-SPL; see Figure 4B; R-Visual: $r = 0.26$, $p = 0.29$; L-Visual: $r = 0.24$, $p = 0.33$). This supports the notion that the correlation between activity level in both SPL during training and subsequent left-hand performance gains is specific to the incongruent training condition. Furthermore, The fact that the R-SPL ROI was obtained whenever visual feedback was added (whether congruent or not) while L-SPL only following the addition of incongruent visual feedback points to functional differences between the two regions with respect to visuomotor adaptation (Mutha et al., 2011).

Neuroimaging: Transfer-Related Network

To further examine the neural correlates of the transfer effect, we performed a functional connectivity analysis (see Experimental Procedures). This method is particularly useful in examining the network level (rather than individual ROIs examined so far) and is based on the dynamics of the signal (rather than the averaged activation of the ROIs collapsed over time). First, we used the activity in L-SPL during RH-LH training as seed. This analysis revealed a network of brain regions that exhibit significant functional connectivity with the L-SPL during training (see Figure 5A). Next, we examined whether across subjects the strength of the functional connectivity between activity in the L-SPL seed region

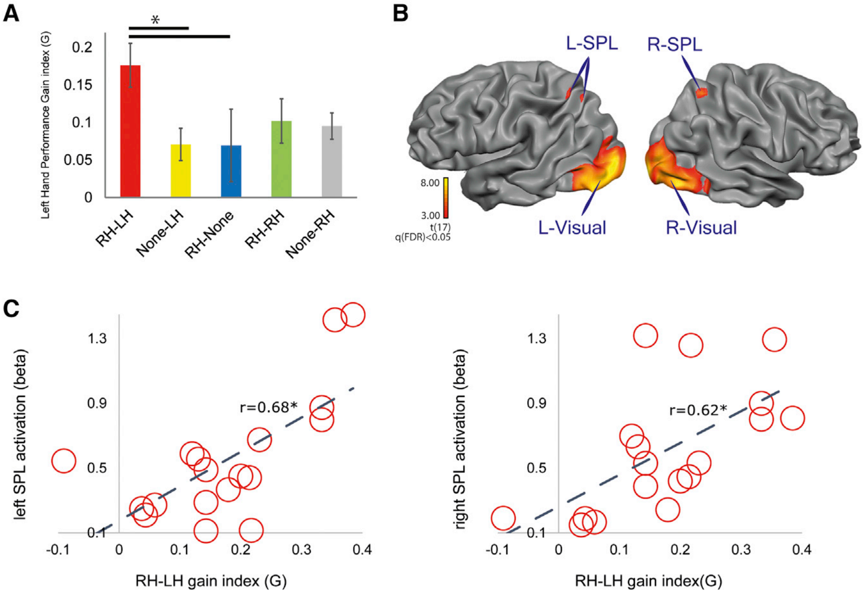

Figure 3. Experiment 3

(A) Behavioral results obtained inside the scanner from the new set of subjects replicated the results from experiment 1. Highest performance gains in the left hand were obtained during RH-LH training ($p < 0.05$). Error bars indicate SEM across subjects.

(B) Regions with enhanced fMRI signal during training with incongruent visual feedback. Multi-subject random effects GLM contrast RH-LH > RH-None ($n = 18$; q(FDR) < 0.05). Right SPL, left SPL and bilateral visual regions were obtained.

(C) The activity in the left SPL (left panel; beta values of the contrast RH-LH > rest during training; see text) and right SPL (right panel) correlated significantly with subsequent left-hand performance gains. Each circle represents one subject.

and activity of individual voxels obtained in the functional connectivity map correlates with their subsequent behavioral changes (see Experimental Procedures). We found that, in a patch of 29 voxels located in the left primary motor cortex (left M1), the connectivity level with L-SPL across individual subjects significantly correlates with subjects' subsequent performance gain in the left hand (q(FDR) < 0.05; see Figure 5B). This shows that subjects exhibiting stronger functional connectivity (cross-talk) between L-SPL and L-M1 during incongruent training also exhibited higher subsequent performance gains in the left hand.

We also performed a similar analysis as above, this time using activity in R-SPL as seed. The functional connectivity map using R-SPL as seed region revealed similar regions to the ones obtained when using L-SPL as seed (86% overlap; see Figure 5B). Interestingly, we found that in a patch of 129 voxels in right occipital gyrus (R-OcG), the strength of the correlation between individual voxel time course and the signal in the R-SPL seed region during RH-LH training co-varied across subjects with their subsequent degree of performance gain in the left hand (q[FDR] < 0.05; see Experimental Procedures). This shows that subjects exhibiting stronger functional connectivity (cross-talk) between R-SPL and R-Visual during incongruent training also exhibited higher subsequent performance gains in the left hand (see Figure 5B). Taken together, this suggests an SPL-based mechanism that plays a significant role in the integration of input received from both the visual and motor cortex during training with incongruent visual feedback. We therefore define the R-SPL, R-OCG, L-SPL, and L-M1 ROIs as transfer-related network for further analysis.

Following the functional connectivity analysis, we examined the relationship between the transfer-related network and regions that are active during right/left-hand movement. To this end, for each subject we performed a GLM analysis on the pre-training evaluation data (in which visual feedback was provided) using two contrasts: $\text{Pre\_Training}_{\text{right\_hand}}$ > rest and $\text{Pre\_Training}_{\text{left\_hand}}$ > rest (see Figure 6A and Experimental Procedures). This analysis yielded 16 regions involved in performance of the finger sequence task. Importantly, in all 18 subjects, the transfer-related network (defined independently based on the group-level correlation with behavior) was located inside these activation regions. Next, subjects were split into two equal groups (median split) according to left-hand performance gains: high learners (subjects with the highest left-hand performance gain following RH-LH training) and low learners. We performed a functional dissimilarity analysis in which functional distances between all 16 regions during RH-LH training were visualized in two dimensions using multi-dimensional scaling for the two groups separately (Figure 6B; see Experimental Procedures). The total functional distance of transfer-related regions (i.e., sum of all distances between transfer-related ROIs) was significantly lower in high versus low learners ($p < 0.05$; equal variance t test). Furthermore, the connectivity level between these regions exhibited a significant inverse correlation with subsequent behavioral changes in left-hand performance (Figure 6C; $p < 0.05$). In other words, subjects with smaller functional distance within the transfer-related network during incongruent training had higher subsequent left-hand performance gain. A similar analysis on these ROIs using signal time courses during training with congruent visual feedback (RH-RH) did not yield significant difference between the two groups ($p = 0.23$; equal variance t test). These results demonstrate that connectivity between visuomotor regions that were identified based on the evaluation stage play a significant role in improved intermanual transfer during training with incongruent visual feedback.

DISCUSSION

Our findings demonstrate the power of sensory feedback on intermanual transfer during short-term motor training. Training by passive observation, in which visual input is not controlled by the subject, has been shown to result in increased performance gains in the same hand (Bird and Heyes, 2005; Mattar and Gribble, 2005). Additionally, physical training with one hand

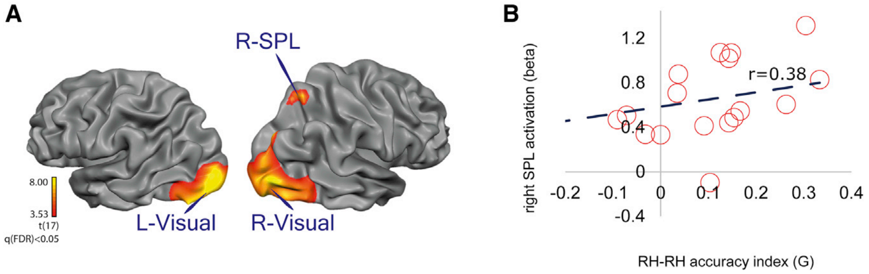

Figure 4. Regions with Enhanced fMRI Signal during Training with Congruent Visual Feedback

(A) Multi-subject random effects GLM contrast RH-RH > RH-None (n = 18; q(FDR) < 0.05). Right SPL and bilateral visual regions exhibited significantly stronger signal in the RH-RH condition. The left SPL was the only region obtained in the RH-LH > RH-None contrast that was not obtained in the current contrast (see Experimental Procedures).

(B) Activity in right SPL was not significantly correlated with subsequent left-hand performance gains (p = 0.11).

has been shown to increase performance gains in the opposite hand (Ruddy and Carson, 2013; Sainburg and Wang, 2002). Here, we show that in the context of intermanual transfer, the combination of physical practice with one hand coupled with incongruent visual feedback that is controlled by the subject yields optimal results in the non-trained hand. Training by observation alone resulted in significantly lower left-hand performance gains irrespective of the identity of the passively observed hand (RH-LH versus None-LH in experiment 1 and versus None-LH and None-RH in experiment 3). Interestingly, physical training with congruent visual feedback (RH-RH) also resulted in lower left-hand performance gains than physical training with incongruent visual feedback (RH-LH; experiments 1 and 3). These results are in line with previous studies which used mirror feedback to demonstrate improved performance of a simple skill in an immobile hand of healthy subjects (Nojima et al., 2012) and accelerated recovery of motor function in an affected hand of stroke patients (Ramachandran and Altschuler, 2009; Sütbeyaz et al., 2007). We extend the research of mirror feedback to the context of motor learning and demonstrate how voluntary control of incongruent visual input during short-term training can enhance intermanual transfer.

Our novel experimental setup allowed us to further show that the addition of passive movement (experiment 2) yields even further transfer gains in the incongruent visual feedback condition (RH-LH-PM). This is consistent with the concept that proprioceptive training improves motor learning in the passively moved effector (Darainy et al., 2013; Vahdat et al., 2014; Wong et al., 2012). It could be argued that, in conditions involving passive left-hand movement, subjects actually performed voluntary bimanual movements. While this cannot be completely ruled out, this alternative explanation is highly unlikely due to several reasons: first, when no visual feedback is provided, we found no significant effect for the addition of passive movement (RH-None versus RH-None-PM); second, there is a slight (non-perceived) delay induced by the device between right-hand movement and yoked left-hand movement (see Experimental Procedures); third, subjective reports of the subjects did not support the feeling of voluntary bimanual movement. Taken together, these results support the notion that passive movement enhances intermanual transfer, such enhancement is sensitive to the type of visual feedback, and most useful in the case of incongruent visual feedback.

Our results are limited to a relatively simple motor task and short-term training. Our training duration is compatible with respect to similar short-term learning studies reported in the literature (Gabitov et al., 2015; Mattar and Gribble, 2005). However, in short-term motor learning, there is a potential for bias from an unspecific arousal effect in the incongruent condition. Therefore, it is possible that across longer training periods, subjects will habituate to the incongruent visual feedback and our reported effects will subside. Therefore, future research is required to examine our effects across multiple training sessions spanning longer time periods (e.g., days or weeks), and their generalization to more complex movements.

Exploiting the current novel approach has the potential for enhancing motor acquisition in clinical patients who exhibit mild to moderate upper extremity hemiparesis. Although in hemiparetic patients direct training of the affected hand has well established effects on rehabilitation (such as constraint-induced movement therapy [Taub et al., 1999; Wolf et al., 2006]), this type of training is very challenging especially in cases where the basic movement capability of the patient is limited (Hoare et al., 2007). We propose to bypass this challenge by combining the VR setup described here with physical training of the non-affected hand. Further studies will be needed to evaluate the effectiveness of such a strategy with respect to direct training of the affected hand.

Finally, we examined possible neural networks underlying the mechanism of enhanced intermanual transfer during training with incongruent visual feedback. We show that subsequent behavioral outcome of such training is reflected by activity in the SPL during training. This is in agreement with studies showing that the parietal cortex and its connection to visual regions are engaged during visuomotor practice (Cross et al., 2009; Culham et al., 2003; Gallivan and Culham, 2015; Krüger et al., 2014; Sakai et al., 2002). We did not detect the ipsilateral primary motor cortex as a correlate to the level of intermanual transfer (Anguera et al., 2007; Farthing et al., 2007; Lee et al., 2010; Perez and Cohen, 2008). A possible explanation might be due to the fact that most physiological studies examining intermanual transfer did not manipulate visual input. We further show that the inter-regional coupling within a network comprising L-SPL and L-M1, and R-SPL and R-visual regions during such training can explain individual differences in subsequent left-hand performance gains. To conclude, our results suggest that, at least when motor skills are concerned, contrary to the popular idiom, my left hand actually *does* know what my right hand is doing, and this knowledge is likely mediated through information conveyed by the SPL.

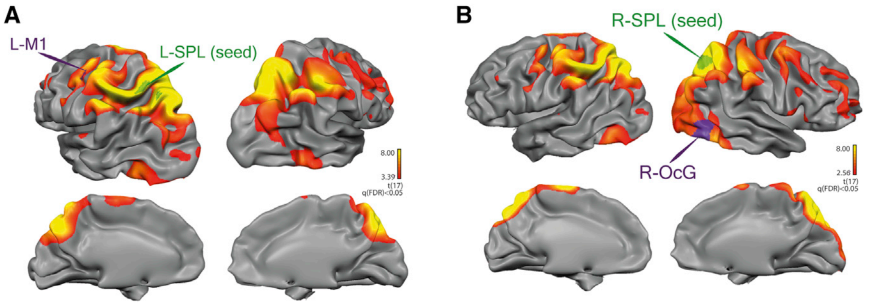

Figure 5. Network Level Analysis

(A) Multi-subject functional connectivity map (n = 18; random effects-q(FDR) < 0.05) using the voxels in left SPL as seed (voxels that correlated with behavior; see Figure 3B) during the RH-LH training stage. The map represents the correlation between activity in the L-SPL seed region and all other voxels. In a patch of 29 voxels located in the left primary motor cortex (left M1; purple voxels), the connectivity level with L-SPL across subjects during RH-LH training significantly correlated with their subsequent left-hand performance gain (q(FDR) < 0.05).

(B) Similar to (A), using the right SPL as seed region (voxels that correlated with behavior; see Figure 3B) during RH-LH training. Across subjects, in a patch of 129 voxels in the right visual area (right occipital gyrus; purple voxels) the degree of connectivity with R-SPL correlated with left-hand performance gains following RH-LH training (q(FDR) < 0.05).

EXPERIMENTAL PROCEDURES

Subjects in all three experiments were healthy and right handed (according to the Edinburgh handedness questionnaire), with normal vision and no reported cognitive deficits or neurological problems. Subjects were naive to the purpose of the study. All the experiments were conducted in accordance with the protocol approved by the Ethics Committee of Tel-Aviv University.

Experiment 1

Eighteen healthy subjects (six females, mean age: 26.3, range: 23–31 years) participated in this study after providing informed consent and were compensated for their participation either by course credit or money (35 New Israeli Shekel [NIS] per hour).

Subjects completed four sessions during which they learned four different sequences of finger movements. Subjects performed the motor task sitting in a chair with their hands forward and palms facing up. Subjects could not see their real hands. Visual feedback of virtual hands was provided through a VR headset used for 3D gaming (Oculus VR, Oculus Rift) (see Figure 1B). Subjects wore motion-sensing MR-compatible gloves (5DT Data Glove Ultra) that allow online monitoring of individual finger flexure in each hand. We also used a head-mounted specialized 3D camera (PLAYSTATION Eye digital camera device) to provide online visual feedback of the real environment. The virtual hands were embedded in a specific location in space and were presented only when subjects looked down toward their real hands (see Movie S1).

In the beginning of each session (Figure 1A), subjects were presented with an instructions slide that depicted two hand illustrations (right/left) with numbered fingers and a five-number sequence underneath, representing the sequence of finger movements to be learned. The instructions slide (12 s) was followed by a pre-training evaluation stage in which baseline performance level of each hand was separately assessed. During the evaluation, subjects performed the required sequence with one hand repeatedly as fast and as accurate as possible for 30 s (hand order right/left was counter-balanced across all sessions). At this stage, online visual feedback consisted of a display of two virtual hands whose finger movements were yoked in real-time to the subjects' actual finger movements. In the following training stage, subjects trained under one of the four following training conditions: RH-RH, RH-LH, RH-None, None-LH (see Table 1; order of training conditions was counter-balanced across subjects). When visual feedback was provided, it included both hands and finger movement was yoked according to the corresponding experimental condition. Following the pre-training evaluation stage, a "Start Training" slide (9 s) cued the subjects to the upcoming training stage in which they performed the sequence of finger movements repeatedly in a self-paced manner. Each training block lasted 15 s followed by 9 s of a yellow blank screen which served as cue for resting period. The training stage consisted of 20 such training blocks. After the training stage, subjects' performance level was re-evaluated as previously in each hand.

Experiment 2

Eighteen healthy subjects (six females, mean age: 25.4, range: 19–36 years) participated in this study and were compensated for their participation (40 NIS per hr). None of the subjects participated in experiment 1.

Subjects completed five sessions in which their task was to learn sequences of finger movements (similar to experiment 1). The experiment setup was identical to experiment 1, with the addition of a specialized motion control apparatus (Rehabit-Tec System; see Figure 2A, Movie S2, and Supplemental Information). Thus, when the hands are strapped to the device and the device is turned on, voluntary right-hand finger movement results in passive yoking of the corresponding left-hand fingers with a slight delay of 191 ms (averaged across fingers). When the device is turned off, right-hand fingers are free to move but left-hand fingers are immobile, and their voluntary movement is impossible.

In each session, subjects trained in one of the following conditions: RH-RH-PM, RH-LH-PM, RH-None-PM, RH-LH, RH-None (see Table 1). During all training conditions, subjects placed their hands inside the device (see Figure 2A). In conditions that did not include passive movements (RH-LH and RH-None), the device was switched off to allow right-hand finger movement while keeping the left hand immobile. Evaluation stages were conducted with palms down outside of the device. The training stages contained ten blocks. Each block lasted 50 s followed by 10 s of rest. Otherwise, the experimental design and subjects' instructions remained similar to experiment 1 (see Figure 1A).

Experiment 3

Eighteen healthy subjects (ten females, mean age: 27.4, range: 22–34 years) participated in this study after providing informed consent. Subjects were recruited according to the standard safety criteria for fMRI studies. One subject also participated in experiment 2. The study conformed to the guidelines of the protocol approved by the Helsinki committee at the Tel-Aviv Sourasky Medical Center. Subjects participated in the experiment either for course credit or money (55 NIS per hr).

During fMRI scans, subjects completed five experimental conditions (five consecutive runs) in which they performed tasks similar to the ones described in experiment 1. We recorded the subjects' finger movements using the MR compatible gloves (same gloves used in experiments 1 and 2) to allow providing online visual feedback of virtual hands presented on a screen. The virtual hands were presented with black background on a screen mounted in front of the subject's eyes in the scanner and viewed through a tilted mirror. Subjects lied supine with their arms to the side of their body and palms facing up. Subjects could not see their real hands during the scans.

Experimental design was identical to experiment 1, with the following training conditions: RH-LH, RH-RH, RH-None, None-LH, and None-RH (see Table 1).

Behavioral Analysis

We used the data from the motion detection gloves to verify that the subjects did not move their fingers during the observational training conditions (None-LH in experiments 1 and 3; None-RH in experiment 3). For further details, see Supplemental Information.

For each subject, we also compared the total amount of self-paced movements performed during training across the different conditions. We used the data from the gloves' sensors, to calculate the amount of movements in each training block (20 blocks in experiments 1 and 3, ten blocks in experiment 2). In all subjects, we found no significant difference between the amount of movements performed during the different training conditions (one-way ANOVA on

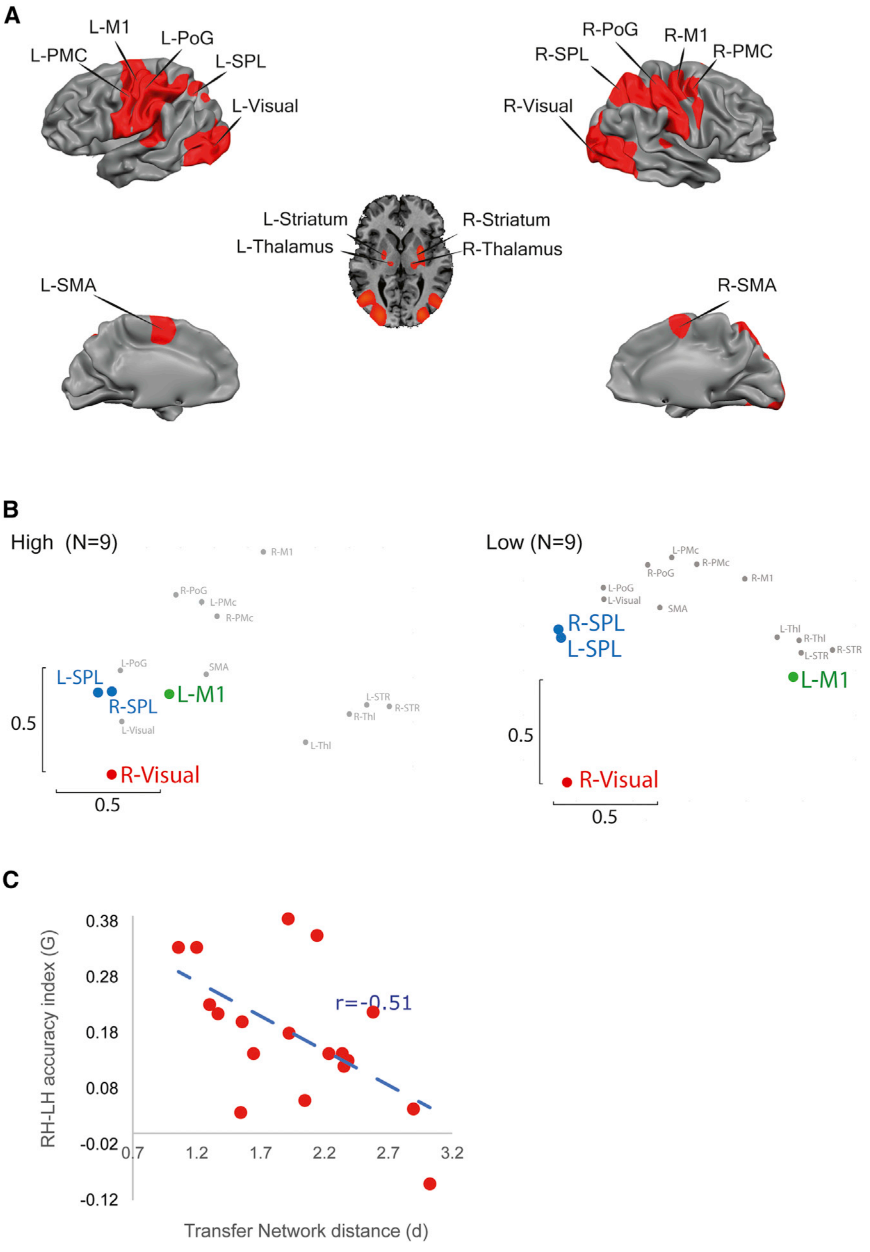

Figure 6. Visuomotor Network Analysis

(A) Random effect multi-subject activation map (n = 18) displaying significant regions obtained from the GLM contrasts RH-RH > rest and LH-LH > rest during the pre-training evaluation. Regions revealed in this contrast include the right and left premotor cortex (R-PMc/L-PMc), primary motor cortex (R-/L-M1), visual cortex (R-Visual/L-Visual), postcentral gyrus (R-PoG/L-PoG), superior parietal lobule (R-SPL/L-SPL), supplementary motor area (R-SMA/L-SMA), and subcortical regions (R-Thalamus/L-Thalamus/R-Striatum/L-Striatum).

(B) Functional distances (between all 16 visuomotor ROIs) in two-dimensional space for good learners (left panel) and bad learners (right panel; see text). The total functional distance within the transfer network (L-SPL, R-SPL, R-visual, and L-M1) is significantly smaller in the high learners than the low learners during training with incongruent visual feedback ($p < 0.05$).

(C) Correlation between functional distance of transfer network during RH-LH training and subsequent left-hand performance gains. Subjects exhibiting smaller functional distances during incongruent RH-LH training exhibited higher subsequent left-hand performance gains ($r = -0.51$, $p < 0.05$). The scatterplot represents summed distance between all ROIs in the transfer network for each subject.

the different training conditions in each experiment; lowest p across all subjects: p = 0.53, p = 0.61, p = 0.34 for experiments 1, 2, and 3, respectively).

Performance Evaluation

In all evaluation stages, we calculated subject's performance (P) by counting the number of correctly performed complete 5-digit sequences within 30 s. Subject's performance gain following training was calculated using the formula below:

$$G = \frac{p_{post\_training} - p_{pre\_training}}{p_{post\_training} + p_{pre\_training}},$$

where $P_{post\_training}/P_{pre\_training}$ corresponds to the subject's performance in the post/pre training evaluation stage. Therefore, a positive G index reflects improvement in performance. We calculate the left-hand performance gain index for each subject, and each experimental condition, allowing us to compare improvement under different motor trainings conditions in each experiment.

fMRI Data Acquisition and Preprocessing

For each subject, blood-oxygenation-level-dependent (BOLD) contrast was obtained using standard protocols described in detail in the Supplemental Information. The acquired data from all subjects underwent preprocessing using the BrainVoyager QX software (version 2.6, Brain Innovation; http://www.brainvoyager.com), as further described in the Supplemental Information.

ROI Analysis

Regions of interest (ROIs; see Figure 3B) were defined at the individual subject level using a general linear model (GLM) contrast to reveal brain regions active during training with congruent and incongruent visual feedback. Next, we examined the correlation between fMRI activity levels across subjects during RH-LH training (Figure 3) or RH-RH training (Figure 4) relative to rest with their corresponding left-hand G value. For further details, see Supplemental Information.

Functional Connectivity Analysis

To examine what brain regions are functionally connected with left or right SPL during RH-LH training, we conducted whole-brain functional connectivity analysis using the activity in left SPL or right SPL as seed regions. Connectivity strength of each voxel in the resulting map with the relevant seed region was examined with respect to left-hand behavioral performance gains (for further details, see Supplemental Information).

Visuomotor Network Analysis

Visuomotor-related regions were defined at the individual subject level by GLM contrasts to reveal brain regions active during the pre-training evaluations collapsed across all sessions of each hand (see multi-subject map in Figure 6A). For further details, see Supplemental Information.

Functional Dissimilarity Analysis

For each subject, we calculated functional distances between the BOLD activation of each pair of visuomotor regions. We then examined these distances in high and low learners. To visualize the distances between all ROIs, we used classical multidimensional scaling (MDS; Figure 6B; [Borg and Groenen, 2005]). For further details, see Supplemental Information.

SUPPLEMENTAL INFORMATION

Supplemental Information includes Supplemental Experimental Procedures, three tables, and two movies and can be found with this article online at http://dx.doi.org/10.1016/j.celrep.2016.11.009.

AUTHOR CONTRIBUTIONS

R.M. and O.O. developed the study concept and designed the experiment; O.O. programmed and collected data; O.O carried out data analysis under supervision from R.M.; O.O. and R.M. wrote the paper and approved the final version.

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REFERENCES

Anguera, J.A., Russell, C.A., Noll, D.C., and Seidler, R.D. (2007). Neural correlates associated with intermanual transfer of sensorimotor adaptation. Brain Res. *1185*, 136–151.

Beets, I.A., Macé, M., Meesen, R.L., Cuypers, K., Levin, O., and Swinnen, S.P. (2012). Active versus passive training of a complex bimanual task: Is prescriptive proprioceptive information sufficient for inducing motor learning? PLoS ONE *7*, e37687.

Bird, G., and Heyes, C. (2005). Effector-dependent learning by observation of a finger movement sequence. J. Exp. Psychol. Hum. Percept. Perform. *31*, 262–275.

Bird, G., Osman, M., Saggerson, A., and Heyes, C. (2005). Sequence learning by action, observation and action observation. Br. J. Psychol. *96*, 371–388.

Borg, I., and Groenen, P.J. (2005). Modern Multidimensional Scaling: Theory and Applications (Springer Science & Business Media).

Brass, M., Bekkering, H., and Prinz, W. (2001). Movement observation affects movement execution in a simple response task. Acta Psychol. (Amst.) *106*, 3–22.

Camus, M., Ragert, P., Vandermeeren, Y., and Cohen, L.G. (2009). Mechanisms controlling motor output to a transfer hand after learning a sequential pinch force skill with the opposite hand. Clin. Neurophysiol. *120*, 1859–1865.

Carroll, T.J., Herbert, R.D., Munn, J., Lee, M., and Gandevia, S.C. (2006). Contralateral effects of unilateral strength training: Evidence and possible mechanisms. J. Appl. Physiol. *101*, 1514–1522.

Criscimagna-Hemminger, S.E., Donchin, O., Gazzaniga, M.S., and Shadmehr, R. (2003). Learned dynamics of reaching movements generalize from dominant to nondominant arm. J. Neurophysiol. *89*, 168–176.

Cross, E.S., Kraemer, D.J., Hamilton, A.F.C., Kelley, W.M., and Grafton, S.T. (2009). Sensitivity of the action observation network to physical and observational learning. Cereb. Cortex *19*, 315–326.

Culham, J.C., Danckert, S.L., DeSouza, J.F., Gati, J.S., Menon, R.S., and Goodale, M.A. (2003). Visually guided grasping produces fMRI activation in dorsal but not ventral stream brain areas. Exp. Brain Res. *153*, 180–189.

Darainy, M., Vahdat, S., and Ostry, D.J. (2013). Perceptual learning in sensorimotor adaptation. J. Neurophysiol. *110*, 2152–2162.

Duque, J., Mazzocchio, R., Stefan, K., Hummel, F., Olivier, E., and Cohen, L.G. (2008). Memory formation in the motor cortex ipsilateral to a training hand. Cereb. Cortex *18*, 1395–1406.

Farthing, J.P., Borowsky, R., Chilibeck, P.D., Binsted, G., and Sarty, G.E. (2007). Neuro-physiological adaptations associated with cross-education of strength. Brain Topogr. *20*, 77–88.

Gabitov, E., Manor, D., and Karni, A. (2015). Patterns of modulation in the activity and connectivity of motor cortex during the repeated generation of movement sequences. J. Cogn. Neurosci. *27*, 736–751.

Gallivan, J.P., and Culham, J.C. (2015). Neural coding within human brain areas involved in actions. Curr. Opin. Neurobiol. *33*, 141–149.

Garry, M.I., Loftus, A., and Summers, J.J. (2005). Mirror, mirror on the wall: Viewing a mirror reflection of unilateral hand movements facilitates ipsilateral M1 excitability. Exp. Brain Res. *163*, 118–122.

Halsband, U., and Lange, R.K. (2006). Motor learning in man: A review of functional and clinical studies. J. Physiol. Paris *99*, 414–424.

Hamzei, F., Läppchen, C.H., Glauche, V., Mader, I., Rijntjes, M., and Weiller, C. (2012). Functional plasticity induced by mirror training: The mirror as the element connecting both hands to one hemisphere. Neurorehabil. Neural Repair *26*, 484–496.

Hendy, A.M., Spittle, M., and Kidgell, D.J. (2012). Cross education and immobilisation: Mechanisms and implications for injury rehabilitation. J. Sci. Med. Sport *15*, 94–101.

Hoare, B.J., Wasiak, J., Imms, C., and Carey, L. (2007). Constraint-induced movement therapy in the treatment of the upper limb in children with hemiplegic cerebral palsy. Cochrane Database Syst. Rev. *2*, CD004149.

Hortobágyi, T., Taylor, J.L., Petersen, N.T., Russell, G., and Gandevia, S.C. (2003). Changes in segmental and motor cortical output with contralateral muscle contractions and altered sensory inputs in humans. J. Neurophysiol. *90*, 2451–2459.

Karni, A., Meyer, G., Rey-Hipolito, C., Jezzard, P., Adams, M.M., Turner, R., and Ungerleider, L.G. (1998). The acquisition of skilled motor performance: Fast and slow experience-driven changes in primary motor cortex. Proc. Natl. Acad. Sci. USA *95*, 861–868.

Kelly, S.W., Burton, A.M., Riedel, B., and Lynch, E. (2003). Sequence learning by action and observation: Evidence for separate mechanisms. Br. J. Psychol. *94*, 355–372.

Krüger, B., Bischoff, M., Blecker, C., Langhanns, C., Kindermann, S., Sauerbier, I., Reiser, M., Stark, R., Munzert, J., and Pilgramm, S. (2014). Parietal and premotor cortices: Activation reflects imitation accuracy during observation, delayed imitation and concurrent imitation. Neuroimage *100*, 39–50.

Lee, M., Hinder, M.R., Gandevia, S.C., and Carroll, T.J. (2010). The ipsilateral motor cortex contributes to cross-limb transfer of performance gains after ballistic motor practice. J. Physiol. *588*, 201–212.

Malfait, N., and Ostry, D.J. (2004). Is interlimb transfer of force-field adaptation a cognitive response to the sudden introduction of load? J. Neurosci. *24*, 8084–8089.

Mattar, A.A., and Gribble, P.L. (2005). Motor learning by observing. Neuron *46*, 153–160.

Muellbacher, W., Facchini, S., Boroojerdi, B., and Hallett, M. (2000). Changes in motor cortex excitability during ipsilateral hand muscle activation in humans. Clin. Neurophysiol. *111*, 344–349.

Mutha, P.K., Sainburg, R.L., and Haaland, K.Y. (2011). Left parietal regions are critical for adaptive visuomotor control. J. Neurosci. *31*, 6972–6981.

Nojima, I., Mima, T., Koganemaru, S., Thabit, M.N., Fukuyama, H., and Kawamata, T. (2012). Human motor plasticity induced by mirror visual feedback. J. Neurosci. *32*, 1293–1300.

Nojima, I., Koganemaru, S., Kawamata, T., Fukuyama, H., and Mima, T. (2015). Action observation with kinesthetic illusion can produce human motor plasticity. Eur. J. Neurosci. *41*, 1614–1623.

Nyberg, L., Eriksson, J., Larsson, A., and Marklund, P. (2006). Learning by doing versus learning by thinking: An fMRI study of motor and mental training. Neuropsychologia *44*, 711–717.

Obayashi, S. (2004). Possible mechanism for transfer of motor skill learning: Implication of the cerebellum. Cerebellum *3*, 204–211.

Ossmy, O., and Mukamel, R. (2016). Activity in superior parietal cortex during training by observation predicts asymmetric learning levels across hands. Sci. Rep. *6*.

Perez, M.A., and Cohen, L.G. (2008). Mechanisms underlying functional changes in the primary motor cortex ipsilateral to an active hand. J. Neurosci. *28*, 5631–5640.

Perez, M.A., Tanaka, S., Wise, S.P., Sadato, N., Tanabe, H.C., Willingham, D.T., and Cohen, L.G. (2007). Neural substrates of intermanual transfer of a newly acquired motor skill. Curr. Biol. *17*, 1896–1902.

Ramachandran, V.S., and Altschuler, E.L. (2009). The use of visual feedback, in particular mirror visual feedback, in restoring brain function. Brain *132*, 1693–1710.

Ruddy, K.L., and Carson, R.G. (2013). Neural pathways mediating cross education of motor function. Front. Hum. Neurosci. *7*, 397.

Sainburg, R.L., and Wang, J. (2002). Interlimb transfer of visuomotor rotations: Independence of direction and final position information. Exp. Brain Res. *145*, 437–447.

Sakai, K., Ramnani, N., and Passingham, R.E. (2002). Learning of sequences of finger movements and timing: Frontal lobe and action-oriented representation. J. Neurophysiol. *88*, 2035–2046.

Scripture, E., Smith, T.L., and Brown, E.M. (1894). On the education of muscular control and power. Stud Yale Psychol Lab *2*, 114–119.

Sigrist, R., Rauter, G., Riener, R., and Wolf, P. (2013). Augmented visual, auditory, haptic, and multimodal feedback in motor learning: A review. Psychon. Bull. Rev. *20*, 21–53.

Sütbeyaz, S., Yavuzer, G., Sezer, N., and Koseoglu, B.F. (2007). Mirror therapy enhances lower-extremity motor recovery and motor functioning after stroke: A randomized controlled trial. Arch. Phys. Med. Rehabil. *88*, 555–559.

Taub, E., Uswatte, G., and Pidikiti, R. (1999). Constraint-Induced Movement Therapy: A new family of techniques with broad application to physical rehabilitation–a clinical review. J. Rehabil. Res. Dev. *36*, 237–251.

Vahdat, S., Darainy, M., and Ostry, D.J. (2014). Structure of plasticity in human sensory and motor networks due to perceptual learning. J. Neurosci. *34*, 2451–2463.

Vogt, S., and Thomaschke, R. (2007). From visuo-motor interactions to imitation learning: Behavioural and brain imaging studies. J. Sports Sci. *25*, 497–517.

Wolf, S.L., Winstein, C.J., Miller, J.P., Taub, E., Uswatte, G., Morris, D., Giuliani, C., Light, K.E., and Nichols-Larsen, D.; EXCITE Investigators (2006). Effect of constraint-induced movement therapy on upper extremity function 3 to 9 months after stroke: The EXCITE randomized clinical trial. JAMA *296*, 2095–2104.

Wolpert, D.M., Diedrichsen, J., and Flanagan, J.R. (2011). Principles of sensorimotor learning. Nat. Rev. Neurosci. *12*, 739–751.

Wong, J.D., Kistemaker, D.A., Chin, A., and Gribble, P.L. (2012). Can proprioceptive training improve motor learning? J. Neurophysiol. *108*, 3313–3321.