

Aus der Abteilung für Funktionelle Bildgebung (Leiter Univ.-Prof. Dr. Martin Lotze)
des Instituts für Diagnostische Radiologie und Neuroradiologie (Direktor Univ.-Prof. Dr.
med. Norbert Hosten) der Universitätsmedizin der Ernst-Moritz-Arndt-Universität
Greifswald



Thema: Modulation motorischen Lernens durch passive sensorische
Stimulation – eine fMRT-Studie zur sensomotorischen Integration bei
Gesunden.

Inauguraldissertation
zur Erlangung des akademischen Grades
eines Doktors der Medizin
der Universitätsmedizin
der Ernst-Moritz-Arndt-Universität
Greifswald
2017

Vorgelegt von: Aija Marie Ladda
geb. am: 26.02.1988
in: Winsen (Luhe)

Wissenschaftlicher Vorstand/Dekan: Univ.-Prof. Dr. rer. nat. Max P. Baur

1. Gutachter: Univ.-Prof. Dr. M. Lotze

2. Gutachter: Univ.-Prof. Dr. B. Sehm

Ort, Raum: Greifswald, Seminarraum B3.49 der Klinik und Poliklinik für Neurologie der
Universitätsmedizin Greifswald

Tag der Disputation: 10.11.2017

Inhaltsverzeichnis

Abbildungsverzeichnis	v
Abkürzungsverzeichnis	v
1 Einleitung	1
1.1 Fragestellung aus Sicht der Grundlagenforschung	1
1.2 Fragestellung aus klinischer Sicht	2
2 Material und Methoden	5
2.1 Probanden	5
2.2 Studienaufbau	6
2.3 Trainingsmethoden	6
2.3.1 Motorisches Training	6
2.3.2 Repetitive Elektrische Stimulation	7
2.4 Motorisches und sensorisches Assessment	8
2.4.1 Motorische Testung trainierter Aufgaben	8
2.4.2 Motorische Testung nicht trainierter Aufgaben	8
2.4.3 Sensorische Testung	8
2.5 Funktionelle Magnetresonanztomografie (fMRT)	9
2.5.1 Grundlagen	9
2.5.2 Messungen	10
2.6 Zusätzliche Analysen	10
2.6.1 Subjektiv empfundene Konzentration und Effekt des Trainings	11
2.6.2 Alltagseinsatz der rechten und der linken Hand	11
2.6.3 Box and Block Test	11
3 Ergebnisse	12
3.1 Sensorik (nur rES-Gruppe)	12
3.2 Motorik	12
3.3 fMRT	13
3.4 Ergebnisse zusätzlicher Analysen	14
3.4.1 Subjektiv empfundene Anstrengung und Konzentration	14
3.4.2 Alltagseinsatz der rechten und der linken Hand	14
3.4.3 Box and Block-Test	14
4 Diskussion	16
5 Literatur	19
6 Zusammenfassung	23

Danksagung	A-1
Publikation A	A-2
Publikation B	A-10

Abbildungsverzeichnis

Abbildung 1:	AFT-Aufgaben	7
Abbildung 2:	Zunahme der funktionellen Aktivierungen der rES-Gruppe im rechten Putamen und linken Cerebellum beim Schreiben mit der linken Hand	13
Abbildung 3:	Ergebnisse des Box and Block Test für die rES-Gruppe	15

Abkürzungsverzeichnis

AFT	Arm-Fähigkeits-Training
BA	Brodmann Area
BBT	Box and Block Test
BOLD	Blood Oxygen Level Dependent
d	Digitus
EEG	Elektroenzephalografie
FWE	Family-wise Error
GOT	Grating Orientation Test
fMRT	funktionelle Magnetresonanztomografie
M1	primär-motorischer Cortex
NHPT	Nine Hole Peg Test
rES	repetitive elektrische Stimulation
RMAT	Roeder Manipulative Aptitude Test
ROI	Region of Interest
S1	primär-somatosensorischer Cortex
S2	sekundär-somatosensorischer Cortex
SMA	supplementär-motorisches Areal
SPL	superiorer Parietallappen
SPM	Statistical Parametric Mapping
tCDS	transkranielle Gleichstromstimulation
TENS	Transkutane elektrische Nervenstimulation
V1	primär-visueller Cortex

1 Einleitung

1.1 Fragestellung aus Sicht der Grundlagenforschung

Auf der Suche nach effektiveren Methoden für sensorische Lernparadigmen wurden Protokolle für passives sensorisches Lernen entwickelt, mit deren Hilfe sich Wahrnehmung und Verhalten des Menschen ohne dessen aktive Mitarbeit beeinflussen lassen. Die Effektivität solcher Protokolle konnte sowohl für visuelle (Beste et al., 2011) als auch für somatosensible (Kalisch et al., 2008) Qualitäten gezeigt werden. Man nimmt an, dass die verwendeten Stimulationsprotokolle die synaptische Übertragung und Effizienz verändern (Beste and Dinse, 2013; Beste et al., 2011). Über bestehende Verbindungen zwischen dem somatosensorischen und dem motorischen Cortex (Jones et al., 1978; Stepniewska et al., 1993; Wu and Kaas, 2003) könnte man durch eine somatosensible Stimulation zusätzlich auch die Erregbarkeit des motorischen Cortex beeinflussen (Ridding et al., 2001), die intracortikale Inhibition dämpfen (Classen et al., 2000) und eine intracortikale Fazilitation fördern (Kobayashi et al., 2003).

Die repetitive Elektrische Stimulation (rES) der Finger ist eine Form des passiven sensorischen Lernens, welche die taktile Wahrnehmung verbessern kann und Plastizitätsprozesse fördert (Godde et al., 2000; Ragert et al., 2008). Bereits die alleinige Anwendung der repetitiven elektrischen Stimulation bei älteren Probanden löste eine Verbesserung der sensomotorischen Leistung aus (Dinse et al., 2006; Kalisch et al., 2008, 2010). In einer anderen Untersuchung der rES korrelierte der individuelle Lernzuwachs hinsichtlich der taktilen Diskriminationsfähigkeit mit der Ausweitung der mit der funktionellen Magnetresonanztomografie (fMRT) gemessenen Repräsentationsareale im primär somatosensorischen Cortex (S1) (Pleger et al., 2003). Man vermutet daher einen Zusammenhang zwischen Veränderungen in frühen sensorischen Arealen und der somatosensorischen Gesamtleistung.

Vor diesem Hintergrund entwickelten wir die Frage, wie sich eine passive somatosensible Stimulation auf ein motorisches Lernparadigma auswirken könnte. Zur Beantwortung dieser Frage muss zunächst erläutert werden, welche Erkenntnisse die Forschung bisher über das motorische Lernen erlangt hat. Wie sieht der zeitliche Ablauf der Aktivitätsänderung der beteiligten Hirnareale aus, und wie hängen Struktur und Funktion im Detail zusammen? Doyon et al. (Doyon et al., 2002) haben am Beispiel einer Fingersequenzaufgabe ein Modell für Lernprozesse im Bereich der Motorik entwickelt. Die

neuronale Repräsentation der Bewegungsprogramme wird in den Anfangsstadien des Lernens im Bereich des cerebellären Cortex angelegt. In frühen Stadien des Lernens wird die Information, die zur Ausführung der Bewegung benötigt wird, zunächst im Nucleus dentatus abgespeichert. Später, in Stadien mit hohem Automatisierungsgrad, wird die Information aus den Basalganglien abgerufen, und die cerebellären Aktivierungen gehen zurück. Das Bewegungsprogramm ist zu diesem Zeitpunkt in ein cortico-striatales Netzwerk eingebunden. Auch Hikosaka et al. (Hikosaka et al., 2002) gehen von einem solchen Modell aus, bei dem, abhängig von der Art der Ausgangskoordinaten der Bewegung (räumliche Koordinaten, oder aber mit Bezug auf die motorischen Einheiten des Körpers), entweder cortico-cerebelläre oder cortico-striatale Schleifen angelegt werden. Über die Bedeutung der Größe der kortikalen Repräsentation des primär-somatosensorischen Cortex (S1) und des primär-motorischen Cortex (M1) in Abhängigkeit von der Dauer der Lerninterventionen gibt es eine kontroverse Debatte. Aufzeichnungen des BOLD-Signals direkt nach dem Training zeigten eine Zunahme der BOLD-Ausdehnung in M1 (Lotze et al., 2003). Im Gegensatz dazu wurde in Langzeitstudien von Trainingseffekten eine fokussierte und ökonomisierte Repräsentation im primär-motorischen Cortex gefunden (Lotze, 2003; Pau et al., 2013). Ähnliches wurde nach einem Training der Wahrnehmung für den visuellen Cortex beschrieben: In den ersten Wochen nahm sowohl die Aktivierung in der zuständigen Unterregion des primär-visuellen Cortex (V1) des trainierten Blickfelds als auch die gemessene Leistung in der Durchführung der Aufgabe zu. Während sich die Performance auf anhaltend hohem Niveau befand, ging die Hirnaktivierung mit der Zeit wieder auf das Ausgangsniveau zurück (Yotsumoto et al., 2008).

1.2 Fragestellung aus klinischer Sicht

Zahlreiche Forschungsgruppen haben insbesondere die motorische Rehabilitation von Schlaganfallpatienten und Trainingsparadigmen für ältere Menschen untersucht. So wissen wir heute nicht nur, welches Training eine gute Erfolgsrate zeigt, sondern wir beginnen auch zu verstehen, wie das Gehirn die notwendigen Umstrukturierungen bewerkstellt. Es konnte vielfach gezeigt werden, dass die funktionelle Kartierung der Hirnleistung weit flexibler ist als gedacht (Thomas et al., 1997; Yotsumoto et al., 2008). Auch die nichtdominante Hemisphäre ist an der Rehabilitation einer geschädigten Funktion der dominanten Hirnhälfte beteiligt (Carey et al., 2006; Feydy et al., 2002).

Da rES wirksam und leicht anzuwenden ist, wird es bei Patienten mit einem Schlaganfall oder einer Hirnverletzung eingesetzt (Kalisch et al., 2008, 2010). Bei der Anwendung über Wochen und längere Zeiträume hinweg konnten positive Auswirkungen auf die taktile Wahrnehmung und die motorische Funktion beobachtet werden (Kattenstroth et al., 2012; Smith et al., 2009). In Studien zu den Effekten der elektrischen Stimulation des Nervus medianus wurde außerdem gezeigt, dass es bei Schlaganfallpatienten zu einer Zunahme der Handkraft kam (Conforto et al., 2002; Dos Santos-Fontes et al., 2013). Eine Kombination von peripherer elektrischer Nervenstimulation und einer eintägigen motorischen Trainingsintervention des Daumens konnte die Trainingseffekte bei Schlaganfallpatienten verstärken (Sawaki et al., 2006).

Unsere Studie folgt der Frage, wie sich der Effekt eines motorischen Trainingsparadigmas durch eine vorbereitende passive sensorische Stimulation mittels rES beeinflussen lässt. Die meisten Studien wiesen Effekte einer elektrischen Stimulation bei Patienten mit neurologischen Erkrankungen nach (Conforto et al., 2007; Sawaki et al., 2006), zu Gesunden wurden hingegen bisher relativ wenige Studien veröffentlicht. Wir nahmen an, dass durch Fazilitierungseffekte auch leistungsstarke junge Erwachsene von einer Kombination aus rES und motorischem Training profitieren würden, mit dem Ergebnis eines schnelleren oder nachhaltigeren Erwerbs motorischer Fertigkeiten. Wir untersuchten sowohl den Leistungszuwachs der motorischen Fähigkeiten als auch der somatosensiblen Diskrimination. Über eine Analyse in vorab definierten Regions of Interest (ROI) prüften wir mittels funktioneller Magnetresonanztomografie (fMRT), ob sich die kortikalen Aktivierungen (sekundär-motorische und -somatosensorische Repräsentationen, die durch motorische Aufgaben aktiviert wurden) im Trainingsverlauf in subkortikal gelegene Areale (anteriore cerebelläre Hemisphäre, Basalganglien) verschieben. Wir verglichen die gemessenen Ergebnisse mit denen einzeln zugeordneter Kontrollprobanden, basierend auf motorischer Ausgangsleistung und demografischen Parametern (Alter, Geschlecht), um festzustellen, ob eine Kombination von somatosensibler Stimulation und motorischen Training gegenüber dem alleinigen motorischen Training zu einem zusätzlichen Effekt führt. Kenntnis der Wirkung und der Funktionsweise der sensomotorischen Integration könnte nicht nur den Eingang der angewandten Methode in die Rehabilitation fördern, sondern öffnet auch Möglichkeiten für die Entwicklung leicht implementierbarer Therapieansätze für stärker betroffene Patienten mit geringer Restfunktion bzw. einer "poor recovery" (vgl. Carey et al., 2006).

Die Ergebnisse der Studie, die dieser Arbeit zugrunde liegt, wurden in zwei Artikeln bei PLOS ONE, Band 9 (Ladda et al., 2014) und in Brain Stimulation, Band 10 (Lotze et al., 2017) veröffentlicht. Eine Kopie beider Publikationen ist am Ende dieser Arbeit beigefügt.

2 Material und Methoden

In diesem Abschnitt werden die Methoden und Materialien, die in dieser Studie zum Einsatz kamen, in abgekürzter Form dargestellt; für detaillierte Informationen insbesondere zu technischen Geräten, Vorverarbeitung und statistischer Auswertung der erhobenen Daten sei an dieser Stelle auf den Methodenteil der zugrundeliegenden Publikationen im Anhang verwiesen.

2.1 Probanden

Wir führten unsere Studie mit insgesamt 30 gesunden jungen Menschen durch, unter Beachtung der Deklaration von Helsinki und mit Zustimmung der Ethikkommission der Medizinischen Fakultät der Universität Greifswald. Die eine Hälfte der Probanden erhielt vor dem motorischen Training jeweils eine elektrische sensorische Stimulation der Fingerspitzen der linken Hand, während in der Kontrollgruppe nur die motorische Trainingsintervention erfolgte. Die Gruppen wiesen in Bezug auf demografische Parameter und die motorische Ausgangsleistung eine homogene Verteilung auf. Es handelte sich um Rechtshänder mit ausgeprägter Dominanz der rechten Hand (Edinburgh Handedness Inventory laterality quotient: $96,1 \pm 4,7$). Außerdem schlossen wir Instrumentalisten von der Studie aus. Keiner der Probanden war von einer neurologischen Störung betroffen, hatte eine bekannte Gefäßerkankung oder nahm eine Dauermedikation ein (hormonelle Kontrazeptiva ausgenommen). Diese Kriterien sollten eine hohe Homogenität innerhalb der Gruppe sicherstellen, insbesondere bezüglich der hemisphäriellen Dominanz. Die interindividuell unterschiedlich ausgeprägten Verbindungen zwischen linker und rechter Hemisphäre werden durch die Dominanz beeinflusst; und die Verteilung der Dominanz verschiedener funktioneller Areale bei unterschiedlicher Händigkeit verhält sich nicht spiegelbildlich zueinander, sodass die Auswertung von Bildgebungsdaten bei gemischten Gruppen Schwierigkeiten bereiten kann (Perry et al., 2001). Darüber hinaus diente die stärkere Naivität der linken Hand bezüglich motorischer Funktionen zur Minimierung eines Deckeneffekts, der im Rahmen des motorischen Trainings auftreten könnte.

2.2 Studienaufbau

Es wurden 30 Rechtshänder untersucht, die ihre linke Hand insgesamt 10 Tage lang motorisch trainierten. Pro Tag wurde etwa 60 Minuten lang trainiert. Die Trainingszeiten wurden dabei mithilfe einer Software dokumentiert (AFT 1.2, Platz, Greifswald; Programmierung durch OLIOID GmbH, Berlin, Deutschland). Der einzelnen Sitzung ging bei 15 Probanden zusätzlich jeweils eine 20-minütige repetitive elektrische Stimulation der Fingerspitzen der linken Hand voraus (rES-Gruppe). Während der Stimulation saßen die Probanden ruhig da, ohne sich im Besonderen auf den Stimulus zu konzentrieren. Ablenkende Aktivitäten wie Lesen oder Fernsehen waren erlaubt.

Als Ergebnisparameter wurde an drei Messzeitpunkten eine motorische Testung durchgeführt. Am ersten und letzten dieser drei Messstage fanden zudem die Messungen im Scanner statt – bei der rES-Gruppe zusätzlich auch eine sensorische Testung.

2.3 Trainingsmethoden

Methoden des motorischen Trainings und der elektrischen Stimulation.

2.3.1 Motorisches Training

Die eingesetzte Trainingsmethode, das Arm-Fähigkeits-Training (AFT), ist in der Rehabilitation von Schlaganfallpatienten bereits etabliert. Es trainiert in acht verschiedenen Aufgaben systematisch überwiegend feinmotorische Fähigkeiten mit jeweils unterschiedlichen Anteilen einer visuellen und somatosensiblen Bewegungskontrolle (s. Abb.1) (Platz et al., 2001). Hierzu zählen schnelle Zielbewegungen, visuelle Folgebewegungen, Arm- und Handstabilität, schnelle isolierte Bewegungen der Finger und die Geschicklichkeit von Hand und Fingern.

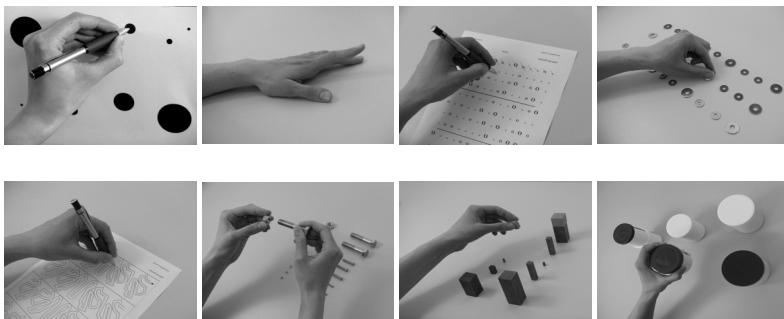


Abbildung 1: AFT-Aufgaben. Von links oben: Zielbewegungen, Fingertapping, Durchstreichen, Scheiben umdrehen, Labyrinth, Schrauben und Muttern, kleine Objekte, große Objekte.

Ziel des Trainings ist eine Reduktion der für die einzelnen Aufgaben benötigten Zeit bei gleichbleibender (oder reduzierter) Fehlerzahl. Über die eingesetzte Software zur Dokumentation konnten die Probanden sich die Ergebnisse ihres Trainings grafisch und numerisch darstellen lassen. An den Tagen der Untersuchungen führten die Probanden das Training unter der Aufsicht des Studienanleiters durch.

2.3.2 Repetitive Elektrische Stimulation

Die 15 Probanden der rES-Gruppe wurden täglich für 20 Minuten an den Fingerspitzen der linken Hand stimuliert. In der Kontrollgruppe wurde keine Stimulation durchgeführt. Zwischen Stimulation und Trainingsbeginn lagen weniger als 20 Minuten. Die Applikation erfolgte über Klebeelektroden am ersten und dritten Fingersegment (Kathode proximal gelegen), die an ein TENS-Gerät angeschlossen wurden. Über ein tragbares Speichermedium spielte das TENS-Gerät eine festgelegte Sequenz einsekündiger Stimulusserien in Intervallen von 5 Sekunden ab (Einzelpulsdauer: 0,2 ms, Rechteckimpuls, Frequenz: 20 Hz). Die Stimulusintensität entsprach dabei der doppelten individuellen Reizschwelle der medianus- und ulnarisversorgten Fingerkuppen; im Mittel $10,8 \pm 1,5$ mA an d1-d3 und $7,8 \pm 0,9$ mA an d4 und d5.

2.4 Motorisches und sensorisches Assessment

2.4.1 Motorische Testung trainierter Aufgaben

Als primärer Ergebnisparameter wurde die Leistung in den AFT-Aufgaben vor und nach dem Training bei allen Probanden für beide Hände einzeln und unverblindet untersucht. Hierzu wurde die Zeit gemessen, welche für die Durchführung der einzelnen Aufgaben benötigt wurde. Die Veränderungen stellten wir jeweils prozentual für das komplette Aufgabenset dar.

2.4.2 Motorische Testung nicht trainierter Aufgaben

Als sekundäre Ergebnisparameter ermittelten wir zum einen die maximale Griffkraft mit einem Vigorimeter (Gebrüder Martin & Co. KG, Tuttlingen, Deutschland). Zur Beurteilung der Feinmotorik wurden außerdem der NHPT und der Roeder-Manipulative-Aptitude-Test (RMAT) durchgeführt. Es handelt sich bei letzterem um ein Testverfahren, welches ursprünglich entwickelt wurde, um die feinmotorischen Fähigkeiten und die Auge-Hand-Koordination potenzieller Arbeitnehmer objektiv zu beurteilen und diese bezüglich der Berufswahl beraten zu können (siehe Roeder Manipulative Aptitude Test Manual, Lafayette Instrument).

2.4.3 Sensorische Testung

Wir verwendeten den Grating Orientation Test (GOT) zur Beurteilung des taktilen Auflösungsvermögens im Bereich der Fingerkuppen. Die Wahrnehmungsschwelle für Druck wurde mittels Frey-Haar-Testung untersucht. Die Testung wurde vor Beginn des Trainings sowie ca. 18-24 Stunden nach Abschluss der Intervention durchgeführt.

2.5 Funktionelle Magnetresonanztomografie (fMRT)

2.5.1 Grundlagen

Zum besseren Verständnis folgt an dieser Stelle in Ergänzung zu den Angaben im Methodenteil der Publikationen ein kurzer Exkurs zur fMRT.

Die funktionellen Magnetresonanztomografie ist eine Untersuchungstechnik, bei der ein erhöhter Energieverbrauch im Gehirn detektiert und lokal zugeordnet werden kann. Sie ermöglicht uns, indirekt auf die Aktivität und Funktion einzelner Hirnareale rückzuschließen. Mittels elektromagnetischer Messung wird die Oxygenierung des Blutfarbstoffes Hämoglobin bestimmt. Die Oxygenierung ist abhängig von der Sauerstoffausschöpfung und somit letztlich vom Energieverbrauch des durchflossenen Hirngewebes. Das aufgenommene Signal bezeichnet man als Blood Oxygenation Level Dependent (BOLD). Da die Änderung des BOLD-Signals von der Flussgeschwindigkeit des Blutes abhängt, ist die zeitliche Auflösung der Signale limitiert und liegt im Bereich von Sekunden. Die räumliche Auflösung der fMRT ist dagegen besonders hoch (< 1cm, u.a. abhängig von der Stärke des Magnetfeldes), z.B. im Vergleich mit der ebenfalls verbreiteten Technik der Elektroenzephalographie (EEG). Die EEG-Technik ist durch eine hohe zeitliche Auflösung im Bereich von Millisekunden gekennzeichnet aber eine geringe räumliche Auflösung.

Um die Wahrscheinlichkeit zu bestimmen, mit der eine gemessene Aktivität der einzelnen Voxel (= Volumenelemente im Bild im Sinne dreidimensionaler Pixel) auch tatsächlich aufgetreten ist, wird zunächst mithilfe des Programms SPM (Statistical Parametric Mapping) pro Voxel eine unabhängige T-Statistik berechnet. Gängig ist dabei die Annahme einer Irrtumswahrscheinlichkeit von 5%. Aufgrund der hohen Gesamtzahl der zu testenden Voxel im Gehirn (das Hirnvolumen beträgt etwa 1400 cm³ (Schrenk, 2008)) kommt es mengenmäßig zu vielen falsch-positiven Aktivierungen (α -Fehler). Die False Discovery Rate (FDR) ermöglicht, dass man ausschließlich die Irrtumswahrscheinlichkeit für diejenigen Voxel berücksichtigt, die als signifikant aktiviert getestet wurden. So verringert sich die Gesamtmenge der falsch-positiven Voxel. Bei der Family-Wise-Error-Korrektur (FWE-Korrektur) wird der Verstärkung eines Bias durch multiple statistische Testungen Rechnung getragen, üblicherweise mittels Anwendung der Bonferroni-Korrektur auf das Ergebnis. Die Summe n aller getesteten Hypothesen H_0 wird verworfen, wenn die Wahrscheinlichkeit p der einzeln getesteten Hypothese kleiner ist als α/n (Wikipedia, 2017). Durch weitere Beschränkung auf die ROI reduziert man die Testung auf

festgelegte Hirnregionen, in welchen man relevante Aktivierungen erwartet oder untersuchen möchte.

2.5.2 Messungen

Die fMRT-Messungen fanden 3-4 Tage vor der Intervention und einen Tag nach der letzten Trainingseinheit statt. Während der Aufzeichnung der Hirnaktivität führten die Probanden im Scanner liegend vier verschiedene motorische Transferaufgaben aus. Zwei dieser Aufgaben ähnelten den im AFT trainierten Bewegungen (visuell geführte Bewegungen: Abschreibaufgabe; und repetitives Fingertapping: Tastendruck nach Zahlenfolge), während die anderen beiden Aufgaben das Erreichen einer vorgegebenen Zielkraft bei wiederholtem Faustschluss sowie die Durchführung des Nine-Hole-Peg-Tests (NHPT) ohne visuelle Kontrolle beinhalteten. Die Aufgaben wurden in einzelnen Durchgängen getrennt mit der rechten und der linken Hand ausgeführt. In der rES-Gruppe gab es zusätzlich zwei Durchgänge mit einer passiven sensorischen Stimulation des linken und des rechten Zeigefingers. Um die spezifische Hirnaktivität während der Aufgaben bzw. der Stimulation gegenüber der Grundaktivität abzugrenzen, erfolgten die Scanuntersuchungen im Blockdesign, bei dem sich Ruhephasen und Aktivität mehrmals während eines Scans abwechseln. Über eine Videoprojektion erhielten die Probanden Start- und Stoppsignale beim Blockwechsel (grüner bzw. blauer Bildschirm), und es wurden die erforderlichen Informationen zur Durchführung der Aufgaben dargestellt (Zahlenfolge als Kodierung für das Drücken korrespondierender Tasten, Rhythmusvorgabe und Feedback-Balken mit Kraft-Ziellinie beim Faustschluss).

Detaillierte Angaben zur Durchführung der Aufgaben und zu den technischen Details der fMRT-Untersuchung sowie die statistische Auswertung finden sich jeweils im Methodenteil der beiden Publikationen im Anhang. Die Ergebnisse der Kontrollgruppe wurden zudem bereits 2015 publiziert (Walz et al., 2015).

2.6 Zusätzliche Analysen

Hierbei handelt es sich um weitere Daten, die erhoben wurden, jedoch nicht mit in die Publikationen eingeflossen sind.

2.6.1 Subjektiv empfundene Konzentration und Effekt des Trainings

Nach der ersten bzw. zweiten Woche baten wir die Probanden um eine Einschätzung mittels visueller Analogskala. Sie sollten angeben, wie hoch ihre Konzentration während des Trainings gewesen war und wie hoch sie den Effekt des bisherigen Trainings einstuften. Der Vergleich der Werte diente zur Abschätzung des Einsatzes der Probanden und möglicher subjektiv wahrgenommener Automatisierungsprozesse.

2.6.2 Alltagseinsatz der rechten und der linken Hand

Die Probanden wurden zu Beginn und Schluss der Studie gebeten die prozentuale Verteilung des Einsatzes der linken und der rechten Hand für insgesamt acht Alltagstätigkeiten anzugeben. Abgefragt wurden folgende Aktivitäten: Zähneputzen, Kämmen, Trinken, Essen mit dem Löffel, Staubwischen, Staubsaugen, Aufräumen und Wäsche waschen/aufhängen.

2.6.3 Box and Block Test

Dieser Test wurde nur in der rES-Gruppe durchgeführt. Mit diesem Test wird eine Bewegung der oberen Extremität untersucht, die grobe Bewegungen des Armes mit einem Präzisionsgriff nach Art des Pinzettengriffs kombiniert. Der Proband sitzt vor einer länglichen Box, die in ein rechtes und ein linkes Kompartiment geteilt ist. Innerhalb von 60 Sekunden müssen so viele Holzklötzchen (2,5 cm x 2,5 cm Kantenlänge) wie möglich einzeln vom ersten Kompartiment auf der Seite der zu testenden Hand über die Trennwand in das zweite Kompartiment befördert werden (Mathiowetz et al., 1985).

3 Ergebnisse

Im Folgenden werden die wesentlichen Ergebnisse der Arbeit in kurzer Form vorgestellt. Der Schwerpunkt liegt hierbei auf dem Vergleich der beiden Studiengruppen. Die Ergebnisse nicht veröffentlichter Analysen werden ebenfalls vorgestellt.

3.1 Sensorik (nur rES-Gruppe)

Die Probanden der rES-Gruppe zeigten nach der Intervention ein erhöhtes taktiles Auflösungsvermögen insbesondere für d2. Darüber hinaus bestand in der Interventionsgruppe eine Korrelation zwischen verbessertem Auflösungsvermögen von d2 und der Verbesserung des Trainingsergebnisses bei der Aufgabe „kleine Objekte“ für die linke Hand. Das unter taktiler Stimulation im MRT aktivierte Areal in Brodmann Area 1 (BA1) war nach der Intervention verkleinert.

3.2 Motorik

Es stellte sich ein insgesamt größerer Trainingserfolg des AAT bei der rES-Gruppe ein (im Mittel 32,9% Verbesserung der Leistung mit der linken Hand) gegenüber der rein motorischen Trainingsintervention ohne vorherige Stimulation (29,5%). Der größte Leistungszuwachs der Stimulationsgruppe gegenüber der Kontrollgruppe war bei der Aufgabe „Fingertapping“ zu verzeichnen. Auch für die rechte Hand zeigte sich in der rES-Gruppe ein vermehrter Leistungszuwachs von 22,0% gegenüber 20,7% in der Kontrollgruppe. Eine schlechte Ausgangsleistung bei der Aufgabe „Schrauben“ konnte als Prädiktor für einen insgesamt besseren Trainingserfolg identifiziert werden. Von den sekundären Ergebnisvariablen zeigte die „Griffkraft“ in der rES-Gruppe für beide Hände eine Kraftzunahme nach dem Training. Die Griffkraft der linken Hand steigerte sich dabei signifikant um 10,0% gegenüber 1,9% in der Kontrollgruppe. Die Schreibgeschwindigkeit der linken Hand nahm in der rES-Gruppe ebenfalls deutlich zu (um 18,6% gegenüber 11,0% bei der Kontrollgruppe). Der Unterschied lag jedoch unterhalb des Signifikanzniveaus und wies eine moderate Effektstärke auf (Cohen's delta: 0,53). Beim NHPT und dem RMAT zeigten sich keine wesentlichen Gruppenunterschiede für die rechte oder die linke Hand (RMAT rechte Hand: $t(28) = 1,29$; n. s.; RMAT linke Hand:

$t(28) = 0,76$; n. s.; NHPT rechte Hand: $t(28) = 0,74$; n. s.; NHPT linke Hand: $t(28) = 0,75$; n. s.). Eine initial niedrige ipsilaterale Aktivierung der sensorischen BA1 prädierte ein besseres Ergebnis bei einer Transferaufgabe mit somatosensorisch geführten Bewegungen (RMAT Aufgabe 1: $r = 0,53$; $P < 0,05$).

3.3 fMRT

Im Direktvergleich der Aktivierungskarten für alle vier linkshändigen Bedingungen der fMRT-Untersuchung zeigte sich sowohl prä minus post als auch post minus prä kein signifikanter Gruppenunterschied.

Die Schreibaufgabe mit der linken Hand nach dem Training löste in der rES-Gruppe eine verstärkte Aktivierung im contralateralen Putamen und im ipsilateralen anterioren Cerebellum aus (Abbildung 2). Mit Ausnahme der Aktivierung des Putamens entsprach dies auch dem Ergebnis der Kontrollgruppe. In beiden Gruppen fand sich bei der Untersuchung der Fingersequenz nach dem Training eine Größenabnahme der aktivierten ROIs im bilateralen primär-sensomotorischen Cortex, dem supplementär-motorischen Areal (SMA), dem dorsolateralen präfrontalen Cortex und dem superioren Parietallappen (SPL). Eine Größenzunahme der Aktivierung fand sich hingegen bei der Griffkraftmodulierung mit der linken Hand. Sie zeigte sich in der rES-Gruppe im Bereich von ipsilateralem Putamen und Pallidum. Während der Durchführung der Fingersequenzaufgabe zeigte sich die Abnahme aktiverter Areale nur für die linke Hand. Bei Durchführung mit der rechten Hand kam es nach der Intervention zu einer vermehrten Aktivierung im Bereich von ipsilateralem Nucleus caudatus und Putamen. In der Kontrollgruppe fand sich die Zunahme der Aktivierung im Bereich der ipsilateralen anterioren cerebellären Hemisphäre, im Vermis und im superioren Parietallappen (BA5). Die Durchführung des NHPT mit der rechten

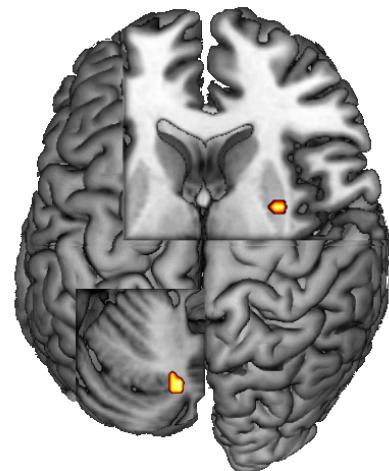


Abbildung 2: Zunahme der funktionellen Aktivierungen der rES-Gruppe im rechten Putamen und linken Cerebellum beim Schreiben mit der linken Hand. Gruppenanalyse für 15 Probanden, Post-minus Prä-Messung; $p < 0,05$; FWE-korrigiert für eine Regions-of-Interest-Analyse.

Hand war in der rES-Gruppe begleitet von einer abnehmenden Aktivität im ipsilateralen Pallidum und einer Aktivitätssteigerung im sekundär-somatosensorischen Cortex (SII).

Bei der Durchführung mit der linken Hand zeigten sich dagegen bei beiden Gruppen keine signifikanten Änderungen.

In beiden Gruppen korrelierte ein positiver Trainingseffekt des gesamten AFT (alle acht Aufgaben) mit einer Zunahme der contralateralen Aktivierung in S1 während der Durchführung der Griffkraftmodulation.

3.4 Ergebnisse zusätzlicher Analysen

3.4.1 Subjektiv empfundene Anstrengung und Konzentration

Die Einschätzung wurde nach der ersten und der zweiten Woche des Trainings erhoben.

In der rES-Gruppe wurde der Effekt des Trainings nach der ersten Woche auf einer visuellen Analogskala auf durchschnittlich $6,4 \pm 0,7$ eingeschätzt; nach der zweiten Woche lag der Mittelwert bei $8,2 \pm 0,6$. Der Wert für die Konzentration wurde in der ersten Woche mit $8,0 \pm 0,8$ angegeben, für die zweite Woche betrug der Mittelwert $8,2 \pm 0,5$.

Die Angaben der Kontrollgruppe unterschieden sich nicht von denen der rES-Gruppe.

3.4.2 Alltagseinsatz der rechten und der linken Hand

Die durchschnittliche Aufgabenteilung zwischen rechter und linker Hand lag für die rES-Gruppe in der Prä-Erhebung bei $89,0 \pm 7,5\%$ bzw. $11,0 \pm 7,5\%$. Nach Abschluss der Studie lag die Verteilung unverändert bei $89,0 \pm 6,5\%$ und $11,0 \pm 6,5\%$. In der Kontrollgruppe war der Alltagseinsatz der Hände vergleichbar.

3.4.3 Box and Block Test

Beim Box and Block Test (BBT) zeigten die Probanden eine signifikante Verbesserung ihrer Ausgangsperformance beider Hände. Die initiale Leistung lag dabei unterhalb der

von Mathiowetz et al. angegebenen Referenzwerte der jeweiligen Altersgruppe (Mathiowetz et al., 1985).

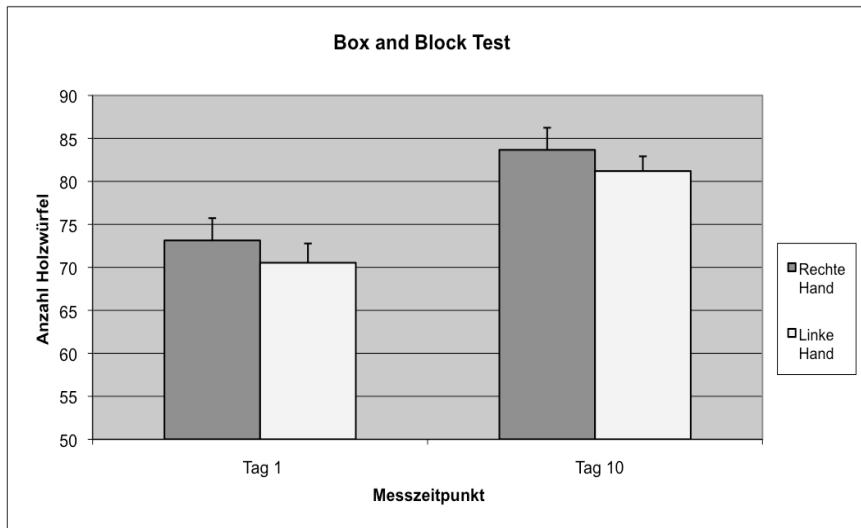


Abbildung 3: Ergebnisse des Box and Block Test für die rES-Gruppe. Verbesserung der Leistung der linken und der rechten Hand mit unverändertem Seitenverhältnis. Die senkrechten Linien geben die Größe des Standardfehlers an.

Die repeated measures Analysis of Variance zeigte für den Faktor Zeit ($F_{(26, 2)} = 40,61; p \leq 0,001$) und Hand ($F_{(13, 1)} = 5,28; p \leq 0,05$) isolierte Effekte (Zeit*Hand: $F_{(26, 2)} = 0,91; n. s.$; zweiseitiger T-Test Zeit: linke Hand $t_{(13)} = -6,06; p \leq 0,001$; linke Hand $t_{(13)} = -6,03; p \leq 0,001$). Zwischen linker und rechter Hand bestand zu keinem Messzeitpunkt ein signifikanter Unterschied der Performance (prä: $t_{(13)} = -1,61; n. s.$; post: $t_{(13)} = -1,36; n. s.$).

4 Diskussion

Bezüglich der taktilen Auflösung konnten wir die Ergebnisse vorhergehender Studien mit dem Grating Orientation Test reproduzieren. Motorisch profitierten die stimulierten Probanden vom Transfereffekt einer zunehmenden Griffkraft, welcher in der Kontrollgruppe nicht eintrat. Den BBT führten wir nur in der rES-Gruppe durch. Es zeigte sich eine Zunahme in der Geschwindigkeit der Durchführung nach dem Training, und beide Hände profitierten in ähnlichem Maße. Da wir feststellten, dass der NHPT in der Testsituation bei Gesunden einige Schwächen aufweist (Wegrutschen der Stifte aus der Mulde), ist zu überlegen, ob in zukünftigen Studien ein anderer motorischer Test eingesetzt werden sollte. Der BBT untersucht im Vergleich gröbere Bewegungen. Der Spitzgriff kommt zwar zum Einsatz; die feine Diskriminationsfähigkeit im Bereich der Fingerkuppen ist jedoch aufgrund der Größe der Holzklötze gegenüber den Anforderungen des NHPT weniger wichtig. Da die Ausgangsleistung der Probanden unterhalb der angegebenen Referenzwerte lag und der Test in der Kontrollgruppe nicht zum Einsatz kam, ist die Einordnung der Ergebnisse schwierig. Insbesondere der durchgehend fehlende Seitenunterschied stellt die Aussagekraft des Tests im vorliegenden Setting in Frage.

Die funktionellen Aktivierungen in der rES-Gruppe wiesen nach dem Training einige Veränderungen auf. Es kam zu einer Ökonomisierung in bilateralen primären und sekundären sensomotorischen Regionen (M1, S1, SMA, SPL) nach dem Training während der Durchführung der Fingersequenz. Für Langzeittraining ist eine solche Ökonomisierung vorbeschrieben; insbesondere an Sequenzaufgaben wurde sie häufig untersucht (Dayan and Cohen, 2011; Shadmehr and Krakauer, 2008). Am Beispiel professioneller Musiker gegenüber Nichtmusikern (Lotze et al., 2003) und Amateuren (Pau et al., 2013) wurde ebenfalls eine Abnahme der motorischen Aktivierung von SMA, prämotorischem Cortex und ipsilateralem M1 bei der Durchführung verschieden komplexer Bewegungen nachgewiesen. Beim Faustschluss stellten wir eine Zunahme der Aktivität im Putamen und im ipsilateralen Kleinhirn fest. Beim Schreiben mit der linken Hand zeigte sich die Zunahme der Aktivierung im contralateralen Putamen. Diese Ergebnisse stützen ebenfalls die bisherige Datenlage zur Veränderung der Aktivierung durch Langzeitlernen (Dayan and Cohen, 2011; Tzvi et al., 2015). Die cerebellären Aktivierungen betrafen dabei sowohl anteriore als auch posteriore Anteile, zudem waren anteriore Anteile des Vermis beteiligt. Für letztere Aktivierung vermuten wir eine Beteiligung bei der Erzeugung einer rhythmischen Komponente des Schreibens (Penhune et al., 1998). Der in beiden Gruppen

beobachtete positive Trainingseffekt im AFT korrelierte mit einer Aktivierungszunahme im contralateralen S1-Areal während der Griffkraftmodulation. Unsere Ergebnisse legen nahe, dass die Verarbeitung sensomotorischer Informationen beider Hände durch ein unilaterales motorisches Training mit einer vorausgehenden somatosensorischen Stimulation modifiziert werden kann. Diese Veränderungen scheinen dabei durch Areale im Bereich der Basalganglien und des Cerebellums vermittelt zu werden. Nach der Stimulation der linken Hand erwarteten wir eine Beeinflussung der Leistung durch eine erhöhte Erregbarkeit der rechten Hemisphäre. Bei der Schreibaufgabe wurden jedoch weiterhin Ressourcen der dominanten linken Hemisphäre rekrutiert. Patienten mit niedriger Ausgangsleistung z.B. aufgrund einer Hirnschädigung könnten von dem Effekt profitieren, wenn sie beim Training auf überlernte Bewegungsmuster der gesunden Hirnhälfte zurückgreifen können. In der Stimulationsgruppe war der Transfereffekt stärker ausgeprägt, die Griffkraft nahm zu. Wir gehen daher von einem generalisierten Lerneffekt für diese Gruppe aus. Die Aktivierung von S1 hingegen nahm in beiden Gruppen zu. Dieses Areal ist von hoher Relevanz für das motorische Lernen. Der Effekt könnte durch eine Beeinflussung der Erregbarkeit der Verbindung zwischen S1 und M1 vermittelt werden (Veldman et al., 2015). S1 wird durch den prämotorischen Cortex kontrolliert (Manita et al., 2015). Ein somatosensibles Defizit beeinträchtigt in der Gegenrichtung über diese Verbindung z.B. die motorische Rehabilitation. In Resting-State-Analysen korrelierten die kortikale Aktivität und Synchronizität positiv mit dem taktilen Diskriminationsvermögen im Bereich der contralateralen Hand. Nach Anwendung von rES zeigte sich eine verbesserte funktionelle Konnektivität zwischen S1 und M1 (Freyer et al., 2012) und die kortikale Erregbarkeit war nach repetitivem taktilem Training gesteigert (Schabrun et al., 2012). Darüber hinaus wird eine Steigerung der Erregbarkeit von M1 in Abhängigkeit von sensorischen Afferenzen über das Cerebellum vermittelt (Popa et al., 2013). Es lässt sich also festhalten, dass Interaktionen zwischen dem motorischen und dem sensorischen Cortex von zentraler Bedeutung sein könnten für die positiven Effekte eines sensorischen Primings der motorischen Leistung. Aufgrund dieser Vermutungen gehen wir davon aus, dass eine gesteigerte Erregbarkeit von S1 zu einer relevanten Verstärkung der primär-sensomotorischen Plastizität führt, und auch zu den beobachteten Trainingseffekten in Bezug auf die somatosensible und motorische Leistung. Die Zunahme der somatosensiblen Leistung könnte insbesondere für die verbesserte Geschwindigkeit des Fingertappings verantwortlich sein, denn diese Aufgabe zeigte die höchste Effektstärke aller AFT-Aufgaben in der rES-Gruppe. Diese Aufgabe wäre darüber hinaus

für den Direktvergleich auch gut im MRT durchführbar und könnte in zukünftigen Studien anstelle des NHPT eingesetzt werden.

Einschränkend ist festzustellen, dass es sich um eine niedrige Probandenzahl handelte, was sich auch im Cohens Delta zeigte. Von der Effektstärke her waren die Ergebnisse vergleichbar mit denen von Studien zur gesteigerten kortikalen Erregbarkeit durch anodale transkranielle Gleichstromstimulation (tCDS). Die rES stellt somit eine Alternative zum anodalen tCDS-Priming dar. Da es sich um Gesunde handelte, war mit einem Deckeneffekt zu rechnen. So zeigte sich kein statistisch relevanter Gruppenunterschied der Aktivierungen im fMRT bei der motorischen Testung. Die geringe statistische Power und die Tatsache, dass die Aufgabensets im Scanner nicht mit dem Training übereinstimmten, dürften hierbei eine Rolle gespielt haben. Auch fanden die Untersuchungen aus organisatorischen Gründen mit einer gewissen Latenz zum Training statt, sodass mittelfristige passagere Veränderungen nicht in die Ergebnisse mit einflossen. Kleine Unterschiede zwischen den Gruppen waren so möglicherweise durch das vorliegende Studiendesign nicht detektierbar. Zudem sind Veränderungen beim Langzeit-Training gegenüber kurzfristigen Lerneffekten insgesamt weniger stark ausgeprägt (Dayan and Cohen, 2011; Lotze, 2003), daher sind unter Umständen dann auch die Gruppenunterschiede bei kleinen Probandenzahlen nicht erkennbar. Ein veränderter Alltagseinsatz der trainierten Hand im Sinne eines globalen Transfereffekts ließ sich ebenfalls nicht nachweisen. In Bezug auf das verwendete rES-Protokoll ist zu erwähnen, dass unter Umständen andere Stimulationsprotokolle (Chipchase et al., 2011; Schabrun et al., 2012) noch effektiver sein könnten in Bezug auf die Modulation von sensomotorischer Interaktion mittels Priming. Bisher gibt es jedoch keine direkten Vergleiche der einzelnen Protokolle. Zum Vergleich der Gruppen bildeten wir sogenannte „matched pairs“. Bei den Kontrollprobanden fehlte eine sensorische Scheinstimulation und Untersuchung, und es gab keine Verblindung. Dies schränkt die Aussagekraft des Vergleichs ein. Zudem gab es keine Kontrollgruppe ohne Training. Gewöhnungseffekte (z.B. bei der fMRT-Untersuchung) lassen sich daher nicht von den Untersuchungsergebnissen isolieren (Schneider and Fink, 2013).

5 Literatur

- Beste, C., and Dinse, H.R. (2013). Learning without Training. *Curr. Biol.* 23, R489–R499.
- Beste, C., Wascher, E., Güntürkün, O., and Dinse, H.R. (2011). Improvement and Impairment of Visually Guided Behavior through LTP- and LTD-like Exposure-Based Visual Learning. *Curr. Biol.* 21, 876–882.
- Carey, L.M., Abbott, D.F., Egan, G.F., O’Keefe, G.J., Jackson, G.D., Bernhardt, J., and Donnan, G.A. (2006). Evolution of brain activation with good and poor motor recovery after stroke. *Neurorehabil. Neural Repair* 20, 24–41.
- Chipchase, L.S., Schabrun, S.M., and Hodges, P.W. (2011). Peripheral electrical stimulation to induce cortical plasticity: a systematic review of stimulus parameters. *Clin. Neurophysiol. Off. J. Int. Fed. Clin. Neurophysiol.* 122, 456–463.
- Classen, J., Steinfelder, B., Liepert, J., Stefan, K., Celnik, P., Cohen, L.G., Hess, A., Kunesch, E., Chen, R., Benecke, R., et al. (2000). Cutaneomotor integration in humans is somatotopically organized at various levels of the nervous system and is task dependent. *Exp. Brain Res.* 130, 48–59.
- Conforto, A.B., Kaelin-Lang, A., and Cohen, L.G. (2002). Increase in hand muscle strength of stroke patients after somatosensory stimulation. *Ann. Neurol.* 51, 122–125.
- Conforto, A.B., Cohen, L.G., dos Santos, R.L., Scaff, M., and Marie, S.K.N. (2007). Effects of somatosensory stimulation on motor function in chronic cortico-subcortical strokes. *J. Neurol.* 254, 333–339.
- Dayan, E., and Cohen, L.G. (2011). Neuroplasticity subserving motor skill learning. *Neuron* 72, 443–454.
- Dinse, H.R., Kleibel, N., Kalisch, T., Ragert, P., Wilimzig, C., and Tegenthoff, M. (2006). Tactile coactivation resets age-related decline of human tactile discrimination. *Ann. Neurol.* 60, 88–94.
- Dos Santos-Fontes, R.L., Ferreiro de Andrade, K.N., Sterr, A., and Conforto, A.B. (2013). Home-based nerve stimulation to enhance effects of motor training in patients in the chronic phase after stroke: a proof-of-principle study. *Neurorehabil. Neural Repair* 27, 483–490.
- Doyon, J., Song, A.W., Karni, A., Lalonde, F., Adams, M.M., and Ungerleider, L.G. (2002). Experience-dependent changes in cerebellar contributions to motor sequence learning. *Proc. Natl. Acad. Sci. U. S. A.* 99, 1017–1022.
- Feydy, A., Carlier, R., Roby-Brami, A., Bussel, B., Cazalis, F., Pierot, L., Burnod, Y., and Maier, M.A. (2002). Longitudinal study of motor recovery after stroke: recruitment and focusing of brain activation. *Stroke* 33, 1610–1617.
- Freyer, F., Reinacher, M., Nolte, G., Dinse, H.R., and Ritter, P. (2012). Repetitive tactile stimulation changes resting-state functional connectivity—implications for treatment of sensorimotor decline. *Front. Hum. Neurosci.* 6.
- Godde, B., Stauffenberg, B., Spengler, F., and Dinse, H.R. (2000). Tactile coactivation-induced changes in spatial discrimination performance. *J. Neurosci. Off. J. Soc. Neurosci.* 20, 1597–1604.
- Hikosaka, O., Nakamura, K., Sakai, K., and Nakahara, H. (2002). Central mechanisms of motor skill learning. *Curr. Opin. Neurobiol.* 12, 217–222.

- Jones, E.G., Coulter, J.D., and Hendry, S.H.C. (1978). Intracortical connectivity of architectonic fields in the somatic sensory, motor and parietal cortex of monkeys. *J. Comp. Neurol.* 181, 291–347.
- Kalisch, T., Tegenthoff, M., and Dinse, H.R. (2008). Improvement of sensorimotor functions in old age by passive sensory stimulation. *Clin. Interv. Aging* 3, 673–690.
- Kalisch, T., Tegenthoff, M., and Dinse, H.R. (2010). Repetitive electric stimulation elicits enduring improvement of sensorimotor performance in seniors. *Neural Plast.* 2010, 690531.
- Kattenstroth, J.-C., Kalisch, T., Peters, S., Tegenthoff, M., and Dinse, H.R. (2012). Long-term sensory stimulation therapy improves hand function and restores cortical responsiveness in patients with chronic cerebral lesions. Three single case studies. *Front. Hum. Neurosci.* 6, 244.
- Kobayashi, M., Ng, J., Théoret, H., and Pascual-Leone, A. (2003). Modulation of intracortical neuronal circuits in human hand motor area by digit stimulation. *Exp. Brain Res.* 149, 1–8.
- Ladda, A.M., Pfannmoeller, J.P., Kalisch, T., Roschka, S., Platz, T., Dinse, H.R., and Lotze, M. (2014). Effects of Combining 2 Weeks of Passive Sensory Stimulation with Active Hand Motor Training in Healthy Adults. *PLoS ONE* 9, e84402.
- Lotze, M. (2003). Motor learning elicited by voluntary drive. *Brain* 126, 866–872.
- Lotze, M., Scheler, G., Tan, H.-R.M., Braun, C., and Birbaumer, N. (2003). The musician's brain: functional imaging of amateurs and professionals during performance and imagery. *NeuroImage* 20, 1817–1829.
- Lotze, M., Ladda, A.M., Roschka, S., Platz, T., and Dinse, H.R. (2017). Priming Hand Motor Training with Repetitive Stimulation of the Fingertips; Performance Gain and Functional Imaging of Training Effects. *Brain Stimulat.* 10, 139–146.
- Manita, S., Suzuki, T., Homma, C., Matsumoto, T., Odagawa, M., Yamada, K., Ota, K., Matsubara, C., Inutsuka, A., Sato, M., et al. (2015). A Top-Down Cortical Circuit for Accurate Sensory Perception. *Neuron* 86, 1304–1316.
- Mathiowetz, V., Volland, G., Kashman, N., and Weber, K. (1985). Adult norms for the Box and Block Test of manual dexterity. *Am. J. Occup. Ther. Off. Publ. Am. Occup. Ther. Assoc.* 39, 386–391.
- Pau, S., Jahn, G., Sakreida, K., Domin, M., and Lotze, M. (2013). Encoding and recall of finger sequences in experienced pianists compared with musically naïve controls: a combined behavioral and functional imaging study. *NeuroImage* 64, 379–387.
- Penhune, V.B., Zattore, R.J., and Evans, A.C. (1998). Cerebellar contributions to motor timing: a PET study of auditory and visual rhythm reproduction. *J. Cogn. Neurosci.* 10, 752–765.
- Perry, R.J., Rosen, H.R., Kramer, J.H., Beer, J.S., Levenson, R.L., and Miller, B.L. (2001). Hemispheric dominance for emotions, empathy and social behaviour: evidence from right and left handers with frontotemporal dementia. *Neurocase* 7, 145–160.
- Platz, T., Winter, T., Müller, N., Pinkowski, C., Eickhof, C., and Mauritz, K.H. (2001). Arm ability training for stroke and traumatic brain injury patients with mild arm paresis: a single-blind, randomized, controlled trial. *Arch. Phys. Med. Rehabil.* 82, 961–968.

- Pleger, B., Foerster, A.F., Ragert, P., Dinse, H.R., Schwenkreis, P., Malin, J.P., Nicolas, V., and Tegenthoff, M. (2003). Functional imaging of perceptual learning in human primary and secondary somatosensory cortex. *Neuron* 40, 643–653.
- Popa, T., Velayudhan, B., Hubesch, C., Pradeep, S., Roze, E., Vidailhet, M., Meunier, S., and Kishore, A. (2013). Cerebellar Processing of Sensory Inputs Primes Motor Cortex Plasticity. *Cereb. Cortex* 23, 305–314.
- Ragert, P., Kalisch, T., Bliem, B., Franzkowiak, S., and Dinse, H.R. (2008). Differential effects of tactile high- and low-frequency stimulation on tactile discrimination in human subjects. *BMC Neurosci.* 9, 9.
- Ridding, M.C., McKay, D.R., Thompson, P.D., and Miles, T.S. (2001). Changes in corticomotor representations induced by prolonged peripheral nerve stimulation in humans. *Clin. Neurophysiol.* 112, 1461–1469.
- Sawaki, L., Wu, C.W.-H., Kaelin-Lang, A., and Cohen, L.G. (2006). Effects of somatosensory stimulation on use-dependent plasticity in chronic stroke. *Stroke* 37, 246–247.
- Schabrun, S.M., Ridding, M.C., Galea, M.P., Hodges, P.W., and Chipchase, L.S. (2012). Primary sensory and motor cortex excitability are co-modulated in response to peripheral electrical nerve stimulation. *PLoS One* 7, e51298.
- Schneider, F., and Fink, G.R. (2013). *Funktionelle MRT in Psychiatrie und Neurologie* (Berlin: Springer).
- Schrenk, Friedemann (2008). *Die Frühzeit des Menschen: Der Weg zum Homo Sapiens*. (Place of publication not identified: C H Beck).
- Shadmehr, R., and Krakauer, J.W. (2008). A computational neuroanatomy for motor control. *Exp. Brain Res.* 185, 359–381.
- Smith, P.S., Dinse, H.R., Kalisch, T., Johnson, M., and Walker-Batson, D. (2009). Effects of Repetitive Electrical Stimulation to Treat Sensory Loss in Persons Poststroke. *Arch. Phys. Med. Rehabil.* 90, 2108–2111.
- Stepniewska, I., Preuss, T.M., and Kaas, J.H. (1993). Architectonics, somatotopic organization, and ipsilateral cortical connections of the primary motor area (M1) of owl monkeys. *J. Comp. Neurol.* 330, 238–271.
- Thomas, C., Altenmüller, E., Marckmann, G., Kahrs, J., and Dichgans, J. (1997). Language processing in aphasia: changes in lateralization patterns during recovery reflect cerebral plasticity in adults. *Electroencephalogr. Clin. Neurophysiol.* 102, 86–97.
- Tzvi, E., Stoldt, A., Witt, K., and Krämer, U.M. (2015). Striatal–cerebellar networks mediate consolidation in a motor sequence learning task: An fMRI study using dynamic causal modelling. *NeuroImage* 122, 52–64.
- Veldman, M.P., Zijdewind, I., Solnik, S., Maffiuletti, N.A., Berghuis, K.M.M., Javet, M., Négyesi, J., and Hortobágyi, T. (2015). Direct and crossed effects of somatosensory electrical stimulation on motor learning and neuronal plasticity in humans. *Eur. J. Appl. Physiol.* 115, 2505–2519.
- Walz, A.D., Doppl, K., Kaza, E., Roschka, S., Platz, T., and Lotze, M. (2015). Changes in cortical, cerebellar and basal ganglia representation after comprehensive long term unilateral hand motor training. *Behav. Brain Res.* 278, 393–403.

Wikipedia (2017). Bonferroni correction. https://en.wikipedia.org/wiki/Bonferroni_correction (Stand: 09. 03. 2017).

Wu, C.W.-H., and Kaas, J.H. (2003). Somatosensory cortex of prosimian Galagos: Physiological recording, cytoarchitecture, and corticocortical connections of anterior parietal cortex and cortex of the lateral sulcus. *J. Comp. Neurol.* 457, 263–292.

Yotsumoto, Y., Watanabe, T., and Sasaki, Y. (2008). Different dynamics of performance and brain activation in the time course of perceptual learning. *Neuron* 57, 827–833.

6 Zusammenfassung

Mit dieser experimentellen Arbeit gelang ein Einblick in die Auswirkungen und das Potenzial eines sensomotorisch kombinierten Lernparadigmas. Dessen Überlegenheit gegenüber einer rein motorischen Intervention zeigt sich in dem insgesamt größeren Leistungszuwachs sowohl der trainierten als auch der untrainierten Hand in der Interventionsgruppe. Wir konnten ein verbessertes taktiles Auflösungsvermögen im stimulierten Bereich nachweisen, welches einherging mit einer abnehmenden kortikalen Aktivierung, was in Analogie zu Studien z.B. an Musikern als Effizienzgewinn in der Informationsverarbeitung zu werten ist. Dass der zusätzliche Trainingsgewinn tatsächlich durch die somatosensible Stimulation verursacht wird, zeigt sich an der Korrelation zwischen der Zunahme des taktilen Auflösungsvermögens und der Verbesserung einer taktil anspruchsvollen Trainingsaufgabe. Ein positiver Effekt zeigte sich auch bei untrainierten Aufgaben und resultierte in einer Zunahme der Griffkraft. Außerdem konnte ein Prädiktor für die Ergebnisse einer somatosensorisch geführten Transferaufgabe identifiziert werden – die niedrige Ausgangsaktivierung in der contralateralen Hirnhälfte zeigte höheres Lernpotenzial an, was auf Erfolge besonders für Schlaganfallpatienten mit schlechter contralateraler Rekrutierung hoffen lässt. Mithilfe der fMRI konnten wir eine Verschiebung der Informationsverarbeitung von kortikal in Richtung Basalganglien und Cerebellum nachweisen, was für einen ausgeprägten Lerneffekt spricht. Die repetitive elektrische Stimulation der Fingerkuppen könnte eine sinnvolle Strategie darstellen, um motorisches Training noch weiter zu verbessern. Die durchschnittliche Leistungssteigerung um 3% macht einen Deckeneffekt bei jungen gesunden Erwachsenen wahrscheinlich. Aufgrund des Potenzials der Intervention bietet es sich an, die Untersuchungen nun auf neurologisch erkrankte und ältere Populationen auszuweiten. Aufgrund der festgestellten Assoziation von Leistungszuwachs und Aktivierungszunahme im primär-somatosensiblen Cortex, sollte das Training bei zukünftigen Studien somatosensible Aspekte stärker berücksichtigen.

Danksagung

Mein Dank gilt den zahlreichen Beteiligten, deren Engagement diese Arbeit möglich gemacht hat. Hierzu zählen neben meinem wunderbaren Doktorvater die übrigen Mitglieder der Abteilung für funktionelle Bildgebung mit der ihr eigenen familiären und hochkonzentrierten Arbeitsatmosphäre, die gutgelaunten MTAs am Scanner, die Kooperationspartner der RUB und der BDH-Klinik, die motivierten Probanden und natürlich meine Familie und meine Freunde, die mir mit Geduld, Interesse und vollster Zuversicht eine stete Stütze waren.

Effects of Combining 2 Weeks of Passive Sensory Stimulation with Active Hand Motor Training in Healthy Adults

Aija Marie Ladda¹, Joerg Peter Pfannmoeller¹, Tobias Kalisch², Sybille Roschka³, Thomas Platz³, Hubert R. Dinse², Martin Lotze^{1*}

1 Functional Imaging Unit, Center for Diagnostic Radiology, University of Greifswald, Greifswald, Germany, **2** Neural Plasticity Lab, Institute for Neuroinformatics, Ruhr-University Bochum, Bochum, Germany, **3** BDH-Klinik Greifswald, Neurorehabilitation Centre and Spinal Cord Injury Unit, University of Greifswald, Greifswald, Germany

Abstract

The gold standard to acquire motor skills is through intensive training and practicing. Recent studies have demonstrated that behavioral gains can also be acquired by mere exposure to repetitive sensory stimulation to drive the plasticity processes. Single application of repetitive electric stimulation (rES) of the fingers has been shown to improve tactile perception in young adults as well as sensorimotor performance in healthy elderly individuals. The combination of repetitive motor training with a preceding rES has not been reported yet. In addition, the impact of such a training on somatosensory tactile and spatial sensitivity as well as on somatosensory cortical activation remains elusive. Therefore, we tested 15 right-handed participants who underwent repetitive electric stimulation of all finger tips of the left hand for 20 minutes prior to one hour of motor training of the left hand over the period of two weeks. Overall, participants substantially improved the motor performance of the left trained hand by 34%, but also showed a relevant transfer to the untrained right hand by 24%. Baseline ipsilateral activation fMRI-magnitude in BA 1 to sensory index finger stimulation predicted training outcome for somatosensory guided movements: those who showed higher ipsilateral activation were those who did profit less from training. Improvement of spatial tactile discrimination was positively associated with gains in pinch grip velocity. Overall, a combination of priming rES and repetitive motor training is capable to induce motor and somatosensory performance increase and representation changes in BA1 in healthy young subjects.

Citation: Ladda AM, Pfannmoeller JP, Kalisch T, Roschka S, Platz T, et al. (2014) Effects of Combining 2 Weeks of Passive Sensory Stimulation with Active Hand Motor Training in Healthy Adults. PLoS ONE 9(1): e84402. doi:10.1371/journal.pone.0084402

Editor: Krish Sathian, Emory University, United States of America

Received October 21, 2013; **Accepted** November 22, 2013; **Published** January 9, 2014

Copyright: © 2014 Ladda et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Funding: A.M. Ladda was supported by a grant from the University of Bochum. M. Lotze, S. Roschka and T. Platz were supported by a grant of the German Research Foundation (Deutsche Forschungsgemeinschaft (LO795/7-1). T. Kalisch and H.R. Dinse acknowledge support by the SFB 874 from the German Research Foundation. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Competing Interests: The authors have declared that no competing interests exist.

* E-mail: martin.lotze@uni-greifswald.de

Introduction

Training-independent sensory learning protocols have been introduced in order to find alternative approaches to motor training to drive changes on human perception and behavior. The effectiveness of such forms of training-independent sensory learning has been demonstrated in different sensory domains. It has been explained by the fact that the stimulation protocols used are capable to alter synaptic transmission and efficacy [1,2]. Repetitive electric stimulation (rES) of the fingers is a form of training-independent sensory learning and has been demonstrated to improve tactile perceptual abilities [3,4] and to drive plasticity processes. Following a single application of rES, individual gains of tactile discrimination were correlated with expansion of blood oxygenation level dependent (BOLD) signals in primary somatosensory cortex (SI) indicating a close link between changes in early sensory areas and overall perceptual performance [5]. rES is similarly effective in elderly individuals thereby resetting the age-related decline of tactile discrimination [6]. Remarkably, rES of the fingers also improved sensorimotor performance in elderly participants [7,8]. Because of its effectiveness and its ease of use, rES is currently applied in patients after stroke or brain lesion.

When applied over weeks or longer, beneficial effects on tactile perception and motor function have been observed [1,9]. Earlier studies on effects of electric stimulation of the median nerve have demonstrated that this procedure increases hand strength in stroke patients [10]. A combination of tactile stimulation with a one-day session of thumb motor training has been described to enhance training effects in stroke patients [11].

While the effectiveness of repetitive sensory stimulation protocols in adult, aged and brain-injury patients is well-documented, so far no study on the combination of rES with motor training has been published in healthy young adults yet. In addition, there is a controversial discussion about associations between representational map size in primary somatosensory cortex (S1) and primary motor cortex (M1) following shorter or longer periods of motor training. Measurements of BOLD -signal immediately after training demonstrated an increase of BOLD-magnitude in M1 [12]. On the contrary, studies measuring long-term training effects observed a more focused and economized representation map in the primary motor cortex [13,14]. A similar observation has been made for the visual cortex (V1) following perceptual training: within the first few weeks of visual training, there were increases

both in activation in the V1 subregion of the trained visual field quadrant and in task performance. But while performance levels remained high, brain activation in the corresponding areas decreased to baseline levels [15]. We hypothesized that through facilitating effects, even high performing young adults would benefit from a combination of rES and motor training, resulting in a more rapid skill acquisition. In addition we wanted to explore the long-term effects on the BOLD-response in relation to tactile discrimination abilities.

Materials and Methods

To investigate the effect of combining rES with active motor training, 15 strongly right-handed young participants underwent a motor training of their left non-dominant hand. We used the left hand to avoid ceiling effects after training which can be expected when testing the dominant hand [16]. We applied the so-called arm ability training (AAT) because it is repetitive, comprehensive, includes concomitant performance measurement and shows good training effects in stroke patients [17,18] and healthy subjects [19]. It trains different abilities such as speed, dexterity, aiming and steadiness. Prior to motor training the finger tips of the left hand were electrically stimulated repeatedly over a period of 20 minutes. Motor outcome was tested with three untrained motor dexterity tests and hand grip force measurement for both hands and assessed using the eight trained motor performance tasks from the AAT. Sensory outcome comprised monofilaments and spatial tactile resolution testing for different fingers of both hands. In addition, we tested the BOLD -response to tactile stimulation of the left index finger pre and post training during fMRI to evaluate changes in index finger representation over the training period.

Participants

We studied 15 right-handed participants each aged 22 to 28 years (age mean = 24.9 ± 2.2 years standard deviation (SD); 7 women). All participants were strongly right-handed (laterality quotient (LQ) = 98.6 ± 3.7 ; range 89–100) according to the Edinburgh Handedness Inventory [20]. None of the participants suffered from any neurological disorder or vascular disease, nor were they on any regular medication (contraceptives excluded).

Participants were recruited via notice boards at the university campus. Any previous or current regular activity in playing musical instruments was considered exclusion criterion for study participation.

Ethics Statement

All participants gave their written and informed consent according to the Declaration of Helsinki, and the study was approved by the ethics committee of the Medical Faculty of the University of Greifswald (BB 126/11).

Experimental schedule

The training period comprised ten consecutive days of training plus one day of rest after the sixth day. On days 1, 2 and 5 training took place in the laboratory in the presence of the instructor. On all the remaining training days participants practiced independently at home. Motor performance was assessed prior to the first training session, on day 5 after the training unit was completed, and one day after the last training session at the end of the second week. Sensory assessment was conducted prior to the first training session and on the day after the last training session as well. fMRI examinations were carried out four days before training started and one day after the last day of training in order to detect long-

term effects only and to avoid short term effects of excitability increase following stimulation [21].

Training

We used the Arm-Ability-Training (AAT) [17] for repetitive and comprehensive motor training of the left non-dominant hand. Participants received detailed instructions on the training method and the documentation software that was being used (AFT 1.2; Platz, Greifswald; Programming by OLIOID GmbH, Berlin, Germany). The AAT comprises eight different tasks (see Figure 1), divided in two training sessions overall approximating 60 minutes a day over two weeks (10 days of training). On the first day participants were taught the correct execution of each task while the instructor was responsible for operating the software which documented performance durations of the tasks during training. From the second day on participants had to operate the software autonomously, i.e. pressing the space key to start and stop the integrated stopwatch, while the instructor supervised correct execution of the tasks.

rES protocol

The participants were stimulated on their left finger tips for 20 minutes/day before they started motor training. The method used was described earlier by Kalisch et al. [7,8]. The rES sequence consisted of stimulus trains of 1 s (single pulse-duration: 0.2 ms (square), frequency: 20 Hz) and inter-train intervals of 5 s. The sequence was played back from a digital storage device that triggered a standard two-channel TENS device (SM2-AKS, Pierenkemper, Germany) via a custom-made input-channel. The pulses were transmitted via adhesive surface electrodes (1 * 4 cm, Pierenkemper, Germany) fixed on the first and third finger-segment (cathode proximal; see Figure 1). Stimulation intensity was adjusted to the twofold sensory threshold separately for median and ulnar nerve innervated fingers resulting in an average initial stimulation intensity of 10.8 ± 1.5 mA on d1–d3 and 7.8 ± 0.9 mA on d4 and d5.

Strength and sensory assessment

Maximum grip force [bar] of both hands was assessed using a vigorimeter (Martin Vigorimeter). Three measures were taken and averaged for each time of measurement.

A Grating Orientation Task (GOT) was used as described by Van Boven et al. [22] to assess the tactile acuity threshold for the area of the fingertip. All fingers of the left hand were tested prior to the first training session and approximately 18–24 hours after completion of the last training session in week 2; additional testing of the right middle finger served as a control condition. Nine different types of hemispherical domes were used for assessment, measuring grating distances of between 0.5 and 3.0 mm. For each size type, 16 trials were performed, and testing started with the greatest distance of gratings. Subjects were asked to close their eyes during the test. Gratings were applied to the distant pad of each finger, either horizontally or vertically oriented, resulting in an indentation of approximately 2 mm and lasting for about 1.5 seconds. The participants were required to make an instant statement about the perceived orientation of the gratings. Testing was aborted when the error-rate of 25% was reached.

For assessment of minimal force-detection threshold, Frey-Hair testing was performed for different localizations: on the fingertips of left d1–d5 and on the dorsum of the left hand, in the radial nerve area. The right hand served as a control condition, testing d1 and d3. Participants were asked to close their eyes and report whenever they felt a sensation on their skin. The filaments were

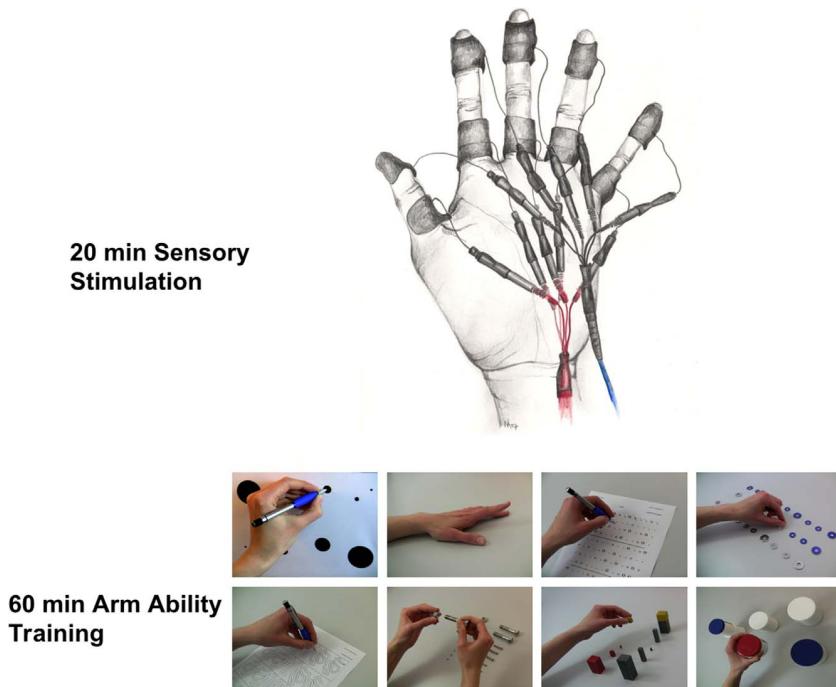


Figure 1. Description of the stimulation and training procedures. Top: Montage of electrodes for repetitive electric stimulation (rES) of the finger tips. Bottom: Eight tasks of the arm ability training (AAT) used for training of the left non-dominant hand: aiming, tapping, crossing, turning coins, labyrinth, bolts and nuts, placing small objects, placing large objects.

doi:10.1371/journal.pone.0084402.g001

pressed against the skin up to three times at a 90° angle until they bowed and were held in place for 1.5 seconds.

Performance testing of trained tasks

On days 1 (prior to the first training session), 5 and 12 (for 5 and 12 after the training session), performance of each of the eight tasks of the standardized Arm-Ability-Training was assessed measuring the time needed [s] to complete the eight AAT-tasks. The trained and untrained hand were tested in a pseudorandomized order. On the first day of testing each participant was allowed an equal minimum of practice to ensure the execution of the task was understood. The tasks covered four different types of movement: Gross force movements (Placing Heavy Objects), sequential finger movements (Tapping) visually guided (Aiming, Crossing Circles, Labyrinth) and somatosensory guided movements (Turning Coins, Nuts And Bolts, Placing Small Objects). To compare different types of movements with regard to the sensory systems predominantly involved in the execution of the particular task we contrasted averages of primarily visually-guided movements with those of non-visually guided movements (somatosensory-guided movements, sequential finger movements). We hypothesized that after application of rES, tests with predominantly visually guided movements would show less improvement compared to the tests that primarily recruit somatosensory resources.

Performance testing of not directly trained tasks

To determine fine motor dexterity the Nine-Hole-PEG-Test was performed with right and left hand, measuring the time needed [s] to take nine pegs from a container mold one by one, insert them into nine holes successively, and then remove the pegs and replace them into the mold again.

The Roeder Manipulative Aptitude Test (RMAT) was used to determine speed and dexterity of arm, hand and finger movements (see Roeder Manipulative Aptitude Test Manual, Lafayette Instrument). Test instructions were given in German language. Performance testing of the first (rods and caps) and the second test (washers and nuts) was conducted. The test involving bilateral hand activity was omitted. Each test was practiced in a standardized way, to allow for stable performance. Each hand was tested separately and a pseudorandomized order was used. For statistical calculation, raw scores of the tests were used.

Statistical analysis

SPSS (V20, IBM Corp., USA) was used for statistical analysis. A 3 TIME (pre, week 1, week 2) and 2 HAND (left, right) repeated measure (RM) ANOVA was conducted to analyze changes of grip force. Somatosensory performance was analyzed by means of 2 TIME RM-ANOVA, separately for each finger. For trained and untrained motor tasks 3 TIME (pre, week 1, week 2) and 2 HAND (left, right) RM-ANOVA were conducted. The direction of significant effect was tested with t-tests corrected for multiple comparisons [23]. For analysis of the AAT-tasks, averages of similar movement type performance were calculated, resulting in the above mentioned categories of trained movements. We performed correlation analyses of the percentual improvement of different somatosensory-guided motor tasks (Turning Coins, Placing Small Objects, Bolts and Nuts, NHPT, RMAT) and the improvement rates of domes discrimination assessment. Based on a ranking of initial performance-level (averaged for all AAT-tasks) we investigated the influence of initial motor performance on the effect of the training method. Ranking in the individual tasks of the AAT was weighted by a score between 1 and 15, with the highest rank (i.e. the fastest performer) receiving 15 points, permitting an

intertask comparison. Adding the scores for individual tasks an overall ranking was conducted for the initial level of AAT-performance. The overall initial performance-level ranking was used as a covariate in the RM-ANOVA evaluating the averaged results of all AAT-tasks. Based on the t-values and considering correlations (*r*-values) of the data, Cohens' *d* was used to calculate the effect size.

MRI Data Acquisition

We used a 3T MRI-scanner (Verio, Siemens, Erlangen, Germany) with a 32 channel head coil. Functional imaging was performed with a standard gradient-echo EPI sequence of 32 transversal slices oriented along the subjects AC-PC plane. In plane resolution was $2 \times 2 \text{ mm}^2$, slice thickness 3 mm and the gap between slices 1 mm. The field of view was $208 \times 208 \text{ mm}^2$ corresponding to an acquisition matrix of 104×104 . Repetition time was 2 s, echo time 23 ms, and the flip angle 90°. Structural imaging was carried out using a sagittal T1-weighted 3D MPRAGE with 176 slices, a spatial resolution of $1 \times 1 \times 1 \text{ mm}^3$ and a gap of 0.5 mm between the slices. The field of view was $250 \times 250 \text{ mm}^2$ corresponding to an acquisition matrix of 256×256 . Repetition time was 1690 ms, echo time 2.52 ms, total acquisition time 3:50 min and the flip angle 9°. In both sequences GRAPPA with a PAT factor of 2 was used.

Tactile Stimulation and Functional Paradigms during MRI

To investigate the changes in the BOLD-response during tactile stimulation, pneumatic stimulus finger clips (MEG International Services Ltd., Coquitlam, Canada) were used to apply tactile stimuli to the subjects' left index finger tips. The stimulators were composed of a support structure and a membrane, with the membrane measuring about 1 cm in diameter. The stimulators were mounted via the support structure and the membrane was actuated by a computer controlled pneumatic valve. During stimulation, pulses with a length of 50 ms and a variable inter-stimulus interval with an average duration of 300 ms were applied for 10 s, resulting in a stimulation frequency of about 3 Hz, which elicited a feeling of pulsating pressure mainly transmitted by Merkel cells [24]. Followed by a rest period of 10 s, this blocked design was repeated 10 times resulting in a total number of 300 stimulations per finger tip. Identical positioning of the stimulators in the pre- and post-examination was assured by photo documentation of the stimulator position in each session. Stimuli application and scanner synchronization were controlled by Presentation software (Neurobehavioral Systems Inc., Albany, USA). We asked the participants to focus attention on the stimuli presented on the index finger.

fMRI Data evaluation

Data were analyzed with the FreeSurfer Analysis Software Suite v5.1 [25]. Since only regions in the cortex were of interest, the surface-based stream was used. The structural scans of each subject were reconstructed automatically and separately for the pre- and the post-examination. The functional scans were evaluated using fs-fast. After motion correction the functional images were co-registered to the subject's anatomical scans using boundary based register and are transformed to fsaverage (MNI305). For removing the effect of individual draining vessels [26] we performed a group average over all subjects, which should minimize the effect of vessels due to the variety in their orientation. Since the somatotopy in Brodmann area 3b needs to be evaluated individually [26] and somatotopic differentiation within this area does not stand normalization procedures, data analysis was restricted to Brodmann area 1 (BA1). BA1 processes tactile shape

recognition [27] and is included in Freesurfer as an anatomical mask [28]. For restricting the somatotopic region to the index finger [29], a tolerance of one standard deviation was included in the label, to account for the variability in the individual functional representation. The contrast of the activation was computed against baseline using a GLM and the group result is smoothed using a Gaussian filter with a 2 mm isotropic kernel. Results were thresholded using a Bonferroni correction for multiple comparisons over the ROI at a significance level of $\alpha = 0.05$.

Results

Grip force and non-trained tasks

For grip force assessment, repeated measures (RM) ANOVA showed a significant influence for the factor TIME ($F_{1,14} = 5.24, P < 0.05$) but not for the factor HAND ($F_{1,14} = 2.31, \text{n.s.}$; no interactions between both factors either: $F_{1,14} = 2.92, \text{n.s.}$). Over both hands, from pre- to post-measurement, grip force increased (two-sided *t*-test: $t_{29} = -3.02, P < 0.01$). Repeated measures ANOVA of the Nine Hole Peg-Test showed significant effects of factors TIME ($F_{1,14} = 11.60, P < 0.001$) and HAND ($F_{1,14} = 6.00, P < 0.05$) without significant interaction. Both hands showed initially similar performance levels ($t_{1,14} = 1.89, \text{n.s.}$) and both improved over time (left: $t_{1,14} = 3.14, P < 0.01$; right: $t_{1,14} = 2.52, P < 0.05$). After the intervention performance of the right hand was significantly faster than left-handed performance ($t_{1,14} = 3.02, P < 0.01$). For the first task of the Roeder Manipulative Aptitude Test similar effects of factors TIME ($F_{1,14} = 61.22, P < 0.001$) and HAND ($F_{1,14} = 79.95, P < 0.001$) were observed, with the right hand at all times performing at a higher level than the left (pre: $t_{1,14} = -7.71, P < 0.001$; post: $t_{1,14} = -7.09, P < 0.001$) and with both hands increasing performance speed over time (left: $t_{1,14} = -5.92, P < 0.001$; right: $t_{1,14} = -5.98, P < 0.001$). For the second task of the Roeder Manipulative Aptitude Test, only the factor TIME was significant ($F_{1,14} = 34.46, P < 0.001$), with the right hand displaying better performance only prior to the intervention ($t_{1,14} = -2.98, P < 0.05$) and both hands improving over time (left hand: $t_{1,14} = -5.60, P < 0.001$; right hand: $t_{1,14} = -2.97, P < 0.001$).

Somatosensory assessment

Tactile acuity increased significantly for left d1 (thumb; RM-ANOVA; d1: $F_{1,14} = 7.78, P < 0.05$), whereas the other digits of the left hand or d3 of the right hand showed no significant changes over time (RM-ANOVA; $F_{1,14} \leq 0.93, \text{n. s.}$). As for the effect size Cohen's *d* was in the middle range for d1 and d3 of the left hand (d1: *d* = 0.64; d3: *d* = 0.53). For the remaining digits of the left hand, Cohen's *d* was low or very low (d2: *d* = 0.07; d4 = 0.46; d5 = 0.17) and the same was found for d3 of the right hand (*d* = 0.11).

Performance of the Frey-Hair test revealed no changes over time (RM-ANOVA $F_{1,14} = 1.35, \text{n. s.}$).

Trained Tasks (AAT-testing)

The gain in performance over time for all AAT-tasks and for either hand displayed high improvement rates (left hand: $34.1 \pm 1.2\%$; right hand: $23.8 \pm 1.0\%$; see Figure 2). Repeated measures ANOVA comparing visually-guided, non-visually-guided tasks revealed significant effects of factors TIME ($F_{2,28} = 270.43, P < 0.001$) and HAND ($F_{1,14} = 67.89, P < 0.001$) on performance time. Significant interactions were found for TIME*HAND ($F_{2,28} = 44.98, P < 0.001$), HAND*TASK ($F_{1,14} = 42.18, P < 0.001$), and TIME*TASK ($F_{2,28} = 5.87, P < 0.01$). Subsequently conducted two-sample *t*-tests revealed significant improvement of performance for all tasks and both hands (visually guided left: $t_{14} = 14.88, P < 0.001$; visually

guided right: $t_{14} = 12.14; P < 0.001$; sensory-guided left: $t_{14} = 16.97; P < 0.001$; sensory-guided right: $t_{14} = 16.59; P < 0.001$; placing heavy objects left: $t_{14} = 10.11; P < 0.001$; placing heavy objects right: $t_{14} = 6.09; P < 0.001$). Initial performance levels for the AAT tasks of the right hand were significantly higher than those of the left hand throughout all the tasks (visually-guided: $t_{14} = 9.87; P < 0.001$; sensory-guided: $t_{14} = 7.82; P < 0.001$; placing heavy objects: $t_{14} = 2.78; P < 0.05$). Performance after two weeks of training showed comparable levels for both hands for sensory-guided movements ($t_{14} = -1.28$; n.s.) and heavy objects ($t_{14} = -1.62$; n.s.), whereas for visually-guided movements the right hand was still faster than the left hand ($t_{14} = 3.47; P < 0.01$).

Cohen's d for these tasks was within significant range for both the left (visually-guided: $d = 0.94$; sensory-guided: $d = 0.95$; heavy objects: $d = 0.92$) and the right hand (visually-guided: $d = 0.93$; sensory-guided: $d = 0.95$; heavy objects: $d = 0.86$).

Correlation Analysis

Increases in discrimination abilities of the left d2 (index finger) as assessed with the domes discrimination task were positively correlated with increases in performance of the AAT-task Placing Small Objects with the left hand ($r = 0.67, P < 0.01$; Figure 3). Furthermore lower baseline performance tended to predict a better outcome after training of screwing small objects ($r = 0.55; P < 0.05$). A lower activation maximum in BA 1 ipsilateral to the stimulated left index finger prior to training predicted a better training outcome for somatosensory driven movements of the trained left hand (Roeder test part 1: $r = 0.53; P < 0.05$).

Representation size in BA1

During the pre-measurement BOLD-response after repetitive tactile stimulation of the left fingertip showed a large representation area in BA1 of the right hemisphere. After 2 weeks, peak activation decreased only moderately. In contrast, the extent of the representational map (map size) decreased considerably (from about 22 to 3 mm²; Figure 3; Table 1) as indicated by a decrease of cluster size by a factor of 7. For the activation maximum we observed a shift in the ROI BA1 of about 3.9 mm which was lower than the effective linear voxel extension of 4 mm and is therefore negligible.

Discussion

Our study showed that even young adult subjects do profit considerably for left hand motor function after a 2-week training, developed for motor training in stroke patients, with a precedent 20 minute period of electric fingertip stimulation. Interestingly, there was a considerable transfer of performance gain between the trained and untrained hand, which improved right hand performance as well thereby hampering a comparison between trained and untrained hand. As expected, rES in combination with motor training had a positive effect on tactile acuity confirming previous findings [3,4,6,7,8]. As a result, subtasks requiring cutaneous input (e.g. placing small objects, bolts and nuts) benefitted from the combined intervention. At the same time we also observed benefits for both trained and untrained tasks and strength of arm and hand-muscles, in line with the findings of Cohen et al. [10]. Notably, for the trained task "placing heavy objects" the left hand showed more improvement than the right, whereas for the untrained (vigorimeter) task both hands profited equally. At a cortical level, the 2 weeks of training with both rES and AAT did not result in an increase of size of fMRI-representation maps of the index finger as one could predict from single applications, but instead representational maps in BA 1 showed a considerable shrinking in map size.

When applied without prior rES in a former study, the arm ability training (AAT) resulted also in a considerable increase of motor function for the left trained hand in young adult healthy participants [19]. These observations suggest that the AAT might be suited for modeling effects of a comprehensive hand training in healthy young adult participants. The observed transfer to the non-trained right hand was high, in line with studies reporting high transfer of trained motor patterns to the untrained hand [30,31], which is independent on the hand side trained, i.e. hand dominance but dependent on the mode of transfer [31] and age of the participants [32], underlining the potential for rehabilitation. Most of the tasks investigated have mirror-image properties and are highly susceptible to intermanual transfer.

Small but significant improvements of dexterity using a peg board task have been reported for young adults following a single session of rES without any motor training [33]. Interestingly in our study we found higher improvement in a peg board task (NHPT)

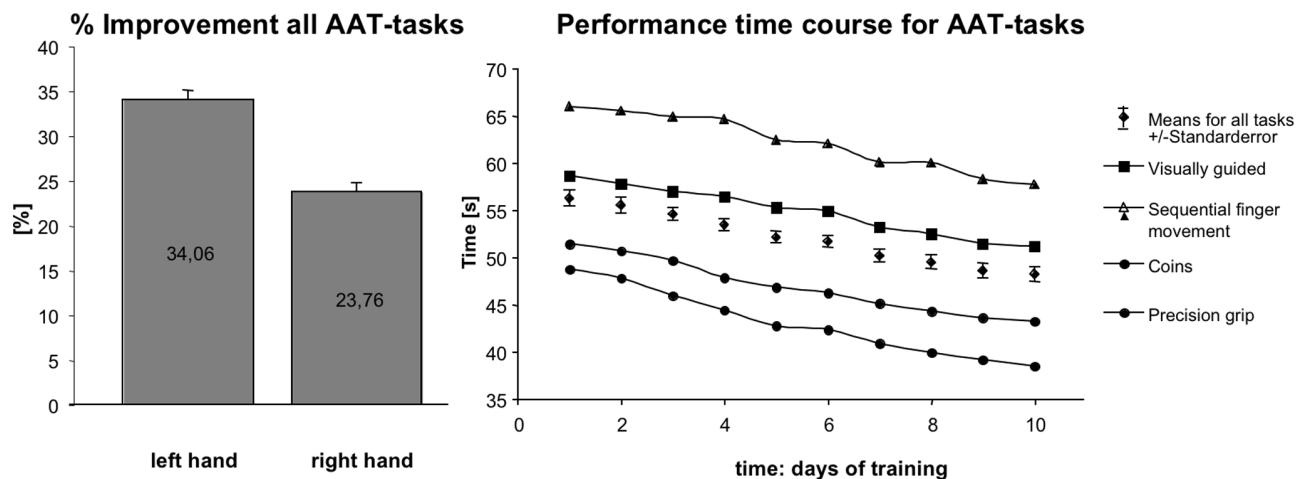


Figure 2. Overview on the performance changes over training time. Left: Average percentual improvement in the AAT-tasks plotted for the trained left and the untrained right hand. Means are provided with standard errors. Right: Detailed absolute increase of performance of the AAT tasks over ten consecutive days for each type of movement trained.
doi:10.1371/journal.pone.0084402.g002

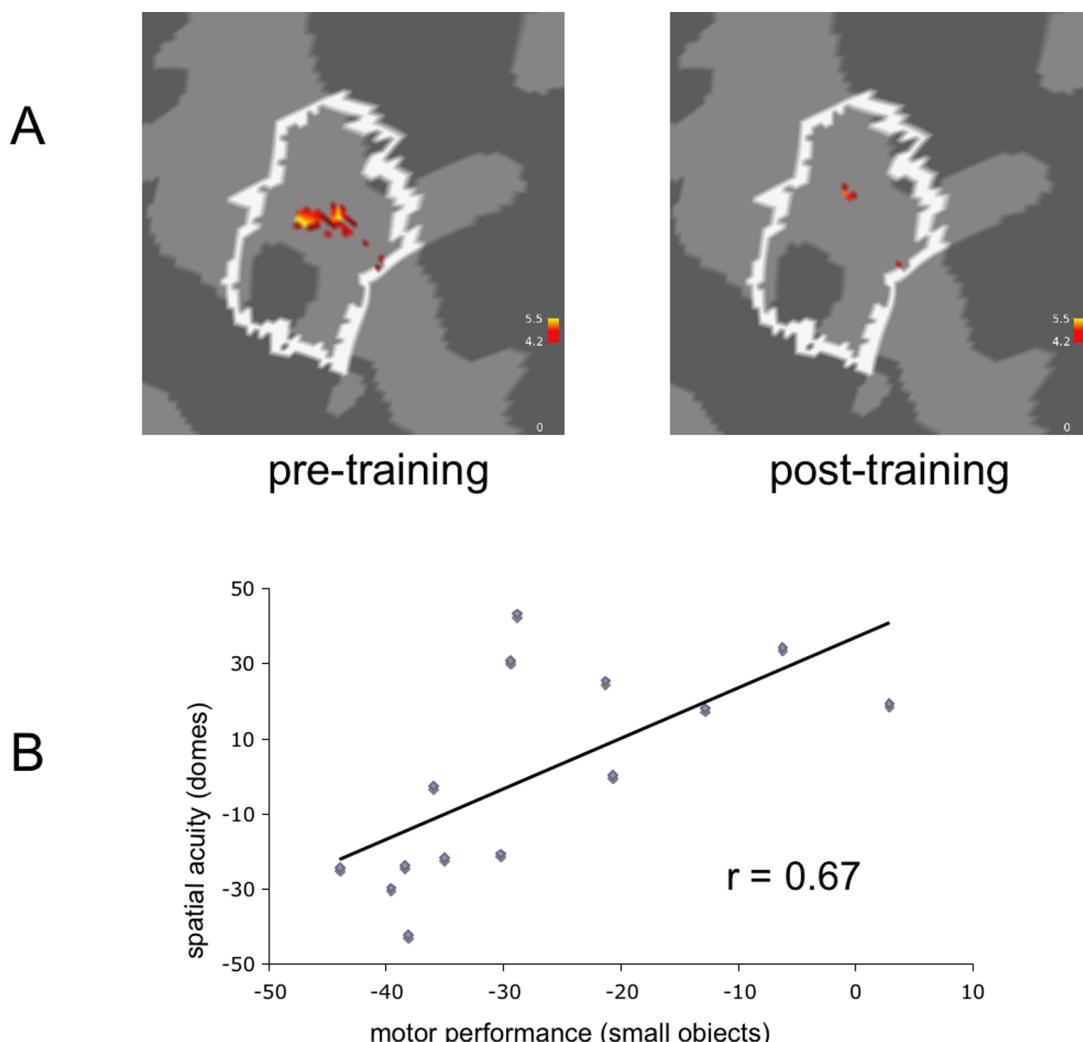


Figure 3. Somatosensory findings on changes over time. A. Visualization of the fMRI-map of the BA 1 activation (ROI indicated with a white line) of the index finger stimulation before (left) and after (right) 2 weeks of intervention (combination of rES and active training). The representational map is decreased in size after two weeks of training although spatial acuity increase was positively associated with increase of pinch grip performance as indicated below. B. The correlation of spatial resolution and motor performance of the AAT task placing small objects was $r=0.67$; $P<0.01$.

doi:10.1371/journal.pone.0084402.g003

for the untrained dominant right hand. To give an explanation for comparable improvement in both hands it would be feasible that the training optimizes recruitment of resources involved in sensorimotor interaction, improving or balancing the communication of the underlying cortical networks. Positive additional effects might be drivable when testing a cohort of less high performing individuals as can be expected in elderly subjects. This

hypothesis would also be supported by the association between baseline-performance and percentual performance gain of a task involving high sensorimotor interaction (nuts and bolts). As for the remaining tests, the left hand seemed to profit particularly in the second RMAT task. The time-crucial component of this task consists in picking up small thin washers laying in a mold in a single layer, demanding high-level sensorimotor-interaction,

Table 1. Parameters of the clusters found in the contralateral (right hemispheric) BA1 for the pre- and the post-examination.

time	maximal activation [t-value]	size [mm^2]	MNI-coordinates			cluster-wise significance
			x	y	z	
pre	5.5	21.8	47.9	-24.6	56	3.6
post	4.9	3.1	47.1	-23.7	59.1	1.2

doi:10.1371/journal.pone.0084402.t001

whereas for the first task, fast movements of the fingers make up a major component while picking up the different items makes up the smaller portion of the task.

rES is capable to increase tactile acuity of the finger tips as has been repeatedly demonstrated for young, adult and elderly participants [3,4,6,7,8]. In the present study, we tested rES in combination with motor training. Therefore, it must remain open, whether the observed increase in acuity of the thumb was due to the additional rES or to the AAT applied alone. During AAT, pinching movements are extensively trained (Figure 1). As a consequence, it is possible that two weeks of AAT affect tactile acuity. Interestingly, pinching movement performance revealed a positive correlation between tactile acuity of the index finger (Figure 3B). A positive association of tactile acuity gain and precise finger pinching movements has been reported after rES in a group of elderly participants [8]. Overall, a combination of AAT with rES might be especially suited for enhancing associations between tactile and motor performance markers, which has to be further investigated in more detail.

In contrast to tactile acuity, touch thresholds as measured with Frey filaments were not altered. This lack of changes of touch threshold has been repeatedly observed in other studies employing rES [33]. It had been argued that the beneficial effects of rES result from changes in synaptic efficacy and synaptic connections. In contrast, touch thresholds seem to reflect predominantly peripheral factors such as mechanoreceptor density and mechanoreceptor composition, which most likely remain unaffected by cortical plasticity processes.

Potential mechanisms of combining rES with active motor training might be related to cortical excitability changes. Cortical excitability is also increased following repetitive transcranial magnetic stimulation (rTMS [34]) or transcranial direct cortical stimulation (TDCS, for a review see [35]). In addition, more focal stimulation strategies through somatosensory electrical nerve stimulation affect cortical excitability [36]. In fact, following rES, SI excitability increases [37], which might enhance effects of a subsequent active motor training.

There is an ongoing debate about cortical economization, habituation effects during repeated measurements and the effect of short- and long-term training on the size of primary representation maps in the somatosensory (S1) and motor (M1) cortex. Very early fMRI studies on repetitive motor training reported a subsequent enlargement of cortical activity in the contralateral M1 during learning (and repetition) of rapid finger movement sequences within the period of a few weeks [38]. Comparably, musicians show an increase of somatosensory representation areas when investigated with magnetoencephalography (MEG) during stimulation of their finger tips in comparison to non-musicians [39]. An enlargement of primary representation areas after training has also been shown specifically for the frequency spectrum of the instrument used in the primary auditory cortex [40]. This long-term training does also result in a more focal representation centered on the contralateral M1 and S1 after years of sensorimotor training [13,41]. However, there are also short-term changes reported for primary motor cortex, where excitability increases within the first 30 minutes after repetitive motor training [21]. For the same experimental situation increased contralateral M1-representations have been demonstrated using fMRI to record BOLD signals [13]. On the other hand, repeated fMRI-measurements without any training have been reported to show habituation effects with decreased representation map size in the contralateral primary sensorimotor cortex [42]. Overall, our data support findings reported for the motor system in stroke patients [43], that an initially high coactivation of the ipsilateral primary

cortex predicts lower performance gain during training and extends it to the somatosensory system.

In our current study we applied elaborate imaging and evaluation techniques using cytoarchitectural masks for defining representational maps. For the ROIs tested, we observed no relevant change of the highest magnitude of activation. Instead, we found a large change in the extent of the representational area. Both observations are in line with the notion of economization of cortical resources after a long-term combined application of rES and active training. It should be noted that our BOLD analysis was restricted to cortical responses in BA 1 leaving out BA 3b. This was based on Schweitzer and colleagues who found that BA 3b representation shows high differences between subjects and thus a normalized evaluation is not possible. Since after normalization S1-response in BA 3b was absent, our present data confirm the occurrence of high interindividual differences. Instead, normalization was assessed as a necessary procedure for eliminating BOLD from vessels which are a relevant problem in evaluating somatotopic representation in individual brains [44]. In future studies we recommend a further increase of spatial resolution and a combination of MRI-angiography and BOLD-imaging to eliminate BOLD from larger vascular origin from analysis of S1-representation sites. Similarly, further studies are needed to obtain analogous information for a group that underwent active motor training only.

These findings are entirely different to those reported earlier using electric source localization [45] or fMRI and BOLD signal recording [5,46] to monitor cortical reorganization following single rES application.

A similar dissociation of reorganization pattern has been made for visual cortex (V1) following perceptual training: within the first few weeks of visual training, there were increases both in activation in the V1 subregion of the trained visual field quadrant and in task performance. But while performance is saturated, brain activation in the corresponding areas decreased to baseline levels [15]. These findings across areas and modalities indicate that there might be distinct temporal phases in which the long-term maintenance of perceptual or behavioral alterations is coded in cortical regions beyond the primary areas. This pattern of changes has been captured by the so-called two-stage model [47,48], according to which plastic changes first develop transiently in early sensory areas, but are then transferred to higher cortical areas, thereby stabilizing the long-term training and learning effects.

Conclusions

Our results point to an increased training effect of sensory stimulation of the finger tips in advance to repetitive motor training at least for those participants who start with lower pre-training motor performance and in particular for tasks that depend on precise somatosensory feedback. This study however, raises several questions concerning underlying cortical mechanisms especially the interactions within the sensorimotor system and the influence of interhemispherical dysbalances, intermanual transfer, long-term effects and application in elderly patients among others which should be addressed in future studies.

Acknowledgments

We thank Christoph Braun and Juergen Dax for substantial support with the sensory stimulation device used in the fMRI-scanner.

Author Contributions

Conceived and designed the experiments: ML AML SR TP. Performed the experiments: ML AML SR RK. Analyzed the data: ML JP AML.

Contributed reagents/materials/analysis tools: JP HD RK TK. Wrote the paper: ML AML HD.

References

- Dinse HR, Gatica Tossi MA, Tegenthoff M, Kalisch T (2011) Sensory stimulation for augmenting perception, sensorimotor behaviour and cognition. In: Markram H SI, editor. *Augmenting cognition*: Epfl Press. pp. 11–39.
- Beste C, Dinse HR (2013) Learning without Training. *Current Biology* 23: R489–R499.
- Godde B, Staufenberg B, Spengler F, Dinse HR (2000) Tactile coactivation-induced changes in spatial discrimination performance. *The Journal of Neuroscience* 20: 1597–1604.
- Ragert P, Kalisch T, Bliem B, Franzkowiak S, Dinse H (2008) Differential effects of tactile high- and low-frequency stimulation on tactile discrimination in human subjects. *BMC Neuroscience* 9: 9.
- Pleger B, Foerster A.-F, Ragert P, Dinse HR, Schwenkreis P, et al. (2003) Functional Imaging of Perceptual Learning in Human Primary and Secondary Somatosensory Cortex. *Neuron* 40: 11.
- Dinse HR, Kleibel N, Kalisch T, Ragert P, Wilimzig C, et al. (2006) Tactile coactivation resets age-related decline of human tactile discrimination. *Annals of Neurology* 60: 88–94.
- Kalisch T, Tegenthoff M, Dinse HR (2008) Improvement of sensorimotor functions in old age by passive sensory stimulation. *Clinical interventions in aging* 3: 673.
- Kalisch T, Tegenthoff M, Dinse HR (2010) Repetitive Electric Stimulation Elicits Enduring Improvement of Sensorimotor Performance in Seniors. *Neural Plasticity* 2010.
- Kattenstroth J-C, Kalisch T, Peters S, Tegenthoff M, Dinse HR (2012) Long-term sensory stimulation therapy improves hand function and restores cortical responsiveness in patients with chronic cerebral lesions. Three single case studies. *Frontiers in human neuroscience* 6: 224.
- Conforto AB, Kaelin-Lang A, Cohen LG (2002) Increase in hand muscle strength of stroke patients after somatosensory stimulation. *Annals of Neurology* 51: 122–125.
- Sawaki L, Wu CW-H, Kaelin-Lang A, Cohen LG (2006) Effects of Somatosensory Stimulation on Use-Dependent Plasticity in Chronic Stroke. *Stroke* 37: 246–247.
- Lotze M, Scheler G, Tan H-R, Braun C, Birbaumer N (2003) The musician's brain: functional imaging of amateurs and professionals during performance and imagery. *Neuroimage* 20: 1817–1829.
- Lotze M, Braun C, Birbaumer N, Anders S, Cohen LG (2003) Motor learning elicited by voluntary drive. *Brain* 126: 866–872.
- Pau S, Jahn G, Sakreida K, Domin M, Lotze M (2012) Encoding and recall of finger sequences in experienced pianists compared to musically naïves: A combined behavioural and functional imaging study. *NeuroImage* 64: 379–387.
- Yotsumoto Y, Watanabe T, Sasaki Y (2008) Different dynamics of performance and brain activation in the time course of perceptual learning. *Neuron* 57: 827–833.
- Petoe MA, Jaque FAM, Byblow WD, Stinear CM (2013) Cutaneous anesthesia of the forearm enhances sensorimotor function of the hand. *Journal of Neurophysiology* 109: 1091–1096.
- Platz T, Winter T, Müller N, Pinkowski C, Eickhof C, et al. (2001) Arm ability training for stroke and traumatic brain injury patients with mild arm paresis: a single-blind, randomized, controlled trial. *Archives of Physical Medicine and Rehabilitation* 82: 961–968.
- Platz T, van Kaick S, Mehrholz J, Leidner O, Eickhof C, et al. (2009) Best conventional therapy versus modular impairment-oriented training for arm paresis after stroke: a single-blind, multicenter randomized controlled trial. *Neurorehabilitation and neural repair* 23: 706–716.
- Platz T, Roschka S, Christel MI, Duecker F, Rothwell JC, et al. (2012) Early stages of motor skill learning and the specific relevance of the cortical motor system—a combined behavioural training and theta burst TMS study. *Restorative Neurology and Neuroscience* 30: 199–211.
- Oldfield RC (1971) The assessment and analysis of handedness: the Edinburgh inventory. *Neuropsychologia* 9: 97–113.
- Classen J, Liepert J, Wise SP, Hallett M, Cohen LG (1998) Rapid Plasticity of Human Cortical Movement Representation Induced by Practice. *Journal of Neurophysiology* 79: 1117–1123.
- Boven RWV, Hamilton RH, Kauffman T, Keenan JP, Pascual-Leone A (2000) Tactile spatial resolution in blind Braille readers. *Neurology* 54: 2230–2236.
- Holm S (1979) A Simple Sequentially Rejective Multiple Test Procedure. *Scandinavian Journal of Statistics* 6: 65–70.
- McGlone F, Reilly D (2010) The cutaneous sensory system. *Neuroscience & Biobehavioral Reviews* 34: 148–159.
- Fischl B (2012) FreeSurfer. *NeuroImage* 62: 774–781.
- Schweisfurth MA, Schweizer R, Frahm J (2011) Functional MRI indicates consistent intra-digit topographic maps in the little but not the index finger within the human primary somatosensory cortex. *NeuroImage* 56: 2138–2143.
- Bodegård A, Geyer S, Grefkes C, Zilles K, Roland PE (2001) Hierarchical Processing of Tactile Shape in the Human Brain. *Neuron* 31: 317–328.
- Fischl B, Rajendran N, Busa E, Augustinack J, Hinds O, et al. (2008) Cortical Folding Patterns and Predicting Cytoarchitecture. *Cerebral Cortex* 18: 1973–1980.
- Weibull A, Björkman A, Hall H, Rosén B, Lundborg G, et al. (2008) Optimizing the mapping of finger areas in primary somatosensory cortex using functional MRI. *Magnetic resonance imaging* 26: 1342–1351.
- Grafton S, Hazeltine E, Ivry R (2002) Motor sequence learning with the nondominant left hand. *Experimental Brain Research* 146: 369–378.
- Kirsch W, Hoffmann J (2010) Asymmetrical intermanual transfer of learning in a sensorimotor task. *Experimental brain research* 202: 927–934.
- Hinder MR, Schmidt MW, Garry MI, Carroll TJ, Summers JJ (2011) Absence of cross-limb transfer of performance gains following ballistic motor practice in older adults. *Journal of Applied Physiology* 110: 166–175.
- Kowalewski R, Kattenstroth J-C, Kalisch T, Dinse HR (2012) Improved acuity and dexterity but unchanged touch and pain thresholds following repetitive sensory stimulation of the fingers. *Neural plasticity* 2012: 974504.
- Stefan K, Gentner R, Zeller D, Dang S, Classen J (2008) Theta-burst stimulation: remote physiological and local behavioral after-effects. *NeuroImage* 40: 265–274.
- Reis J, Fritsch B (2011) Modulation of motor performance and motor learning by transcranial direct current stimulation. *Current opinion in neurology* 24: 590–596.
- Conforto A, Cohen L, Santos R, Scaff M, Marie S (2007) Effects of somatosensory stimulation on motor function in chronic cortico-subcortical strokes. *Journal of Neurology* 254: 333–339.
- Höffken O, Veit M, Knosalla F, Lissek S, Bliem B, et al. (2007) Sustained increase of somatosensory cortex excitability by tactile coactivation studied by paired median nerve stimulation in humans correlates with perceptual gain. *The Journal of Physiology* 584: 463–471.
- Karni A MG, Jezzard P, Adams MM, Turner R, Ungerleider LG (1995) Functional MRI evidence for adult motor cortex plasticity during motor skill learning. *Nature* 377: 155–158.
- Elbert T, Pantev C, Wienbruch C, Rockstroh B, Taub E (1995) Increased Cortical Representation of the Fingers of the Left Hand in String Players. *Science* 270: 305–307.
- Pantev C, Oostenveld R, Engelen A, Ross B, Roberts LE, et al. (1998) Increased auditory cortical representation in musicians. *Nature* 392: 811–814.
- Hund-Georgiadis M, von Cramon DY (1999) Motor-learning-related changes in piano players and non-musicians revealed by functional magnetic-resonance signals. *Experimental Brain Research* 125: 417–425.
- Loubinoux I, Carel C, Alary F, Boulanouar K, Viallard G, et al. (2001) Within-Session and Between-Session Reproducibility of Cerebral Sensorimotor Activation: A Test–Retest Effect Evidenced With Functional Magnetic Resonance Imaging. *Journal of Cerebral Blood Flow & Metabolism* 21: 592–607.
- Dong Y, Dobkin BH, Cen SY, Wu AD, Winstein CJ (2006) Motor cortex activation during treatment may predict therapeutic gains in paretic hand function after stroke. *Stroke* 37: 1552–1555.
- Schweizer R, Voit D, Frahm J (2008) Finger representations in human primary somatosensory cortex as revealed by high-resolution functional MRI of tactile stimulation. *NeuroImage* 42: 28–35.
- Pleger B, Dinse HR, Ragert P, Schwenkreis P, Malin JP, et al. (2001) Shifts in cortical representations predict human discrimination improvement. *Proceedings of the National Academy of Sciences* 98: 12255–12260.
- Hodzic A, Veit R, Karim AA, Erb M, Godde B (2004) Improvement and Decline in Tactile Discrimination Behavior after Cortical Plasticity Induced by Passive Tactile Coactivation. *The Journal of Neuroscience* 24: 442–446.
- Watanabe T, Náñez JE, Koyama S, Mukai I, Liederman J, et al. (2002) Greater plasticity in lower-level than higher-level visual motion processing in a passive perceptual learning task. *Nature neuroscience* 5: 1003–1009.
- Sagi D (2011) Perceptual learning in Vision Research. *Vision research* 51: 1552–1566.



Priming Hand Motor Training with Repetitive Stimulation of the Fingertips; Performance Gain and Functional Imaging of Training Effects



Martin Lotze ^{a,*}, Aija Marie Ladda ^a, Sybille Roschka ^b, Thomas Platz ^b, Hubert R. Dinse ^{c,d}

^a Functional Imaging Unit, Center for Diagnostic Radiology, University of Greifswald, Germany

^b BDH-Klinik Greifswald, Neurorehabilitation centre and Spinal Cord Injury Unit, University of Greifswald, Germany

^c Neural Plasticity Lab, Institute for Neuroinformatics, Ruhr-University Bochum, Germany

^d Department of Neurology, BG University Hospital Bergmannsheil, Ruhr University Bochum, Germany

ARTICLE INFO

Article history:

Received 27 April 2016

Received in revised form 1 October 2016

Accepted 5 October 2016

Available online 6 October 2016

Keywords:

Motor training

Stimulation

Tactile priming

fMRI

Arm ability training

ABSTRACT

Background: Application of repetitive electrical stimulation (rES) of the fingers has been shown to improve tactile perception and sensorimotor performance in healthy individuals.

Objective: To increase motor performance by priming the effects of active motor training (arm ability training; AAT) using rES.

Methods: We compared the performance gain for the training increase of the averaged AAT tasks of both hands in two groups of strongly right-handed healthy volunteers. Functional Magnetic Resonance Imaging (fMRI) before and after AAT was assessed using three tasks for each hand separately: finger sequence tapping, visually guided grip force modulation, and writing. Performance during fMRI was controlled for precision and frequency. A total of 30 participants underwent a two-week unilateral left hand AAT, 15 participants with 20 minutes of rES priming of all fingertips of the trained hand, and 15 participants without rES priming.

Results: rES-primed AAT improved the trained left-hand performance across all training tasks on average by 32.9%, non-primed AAT improved by 29.5%. This gain in AAT performance with rES priming was predominantly driven by an increased finger tapping velocity. Functional imaging showed comparable changes for both training groups over time. Across all participants, improved AAT performance was associated with a higher contralateral primary somatosensory cortex (S1) fMRI activation magnitude during the grip force modulation task.

Conclusions: This study highlights the importance of S1 for hand motor training gain. In addition, it suggests the usage of rES of the fingertips for priming active hand motor training.

© 2016 Elsevier Inc. All rights reserved.

Introduction

Repetitive somatosensory stimulation (rSS) of the fingers, either tactile or electrical, has been demonstrated to drive plasticity processes and to improve tactile perceptual abilities [1]. rSS is capable to reset the age-related decline of tactile discrimination in elderly individuals [2] and has also an impact on motor function [3]. The effectiveness of this method has been demonstrated in patients with chronic cerebral lesions during long (weeks to months) stimulation periods [4], and recently also in patients suffering from neuropathic pain [5]. Both stimulations of two and three hand nerves are comparably capable to improve motor function in healthy young

participants [6]. However, somatosensory electrical stimulation was not capable to increase short term motor training effects in healthy volunteers in an earlier study [7].

Besides improvement of somatosensory abilities, application of rSS also improved motor performance in both healthy adults and elderly individuals [3,8] and patients [4]. However, how rSS affects the motor system remains largely speculative. It is generally believed that the transfer of beneficial effects to sensorimotor behavior elicited by sensory stimulation is based on interconnections between the somatosensory and motor cortices [9–11]. These interconnections are assumed to elicit a cortical reorganization in the primary motor cortex after stimulation, resulting in increased excitability of the motor cortical representations [12], in intracortical facilitation [13], and in a decrease in intracortical inhibition [14]. On the other hand, accurate sensory perception requires recurrent SI activation from the secondary motor cortex [15].

* Corresponding author. Fax: +49 3834 866898.

E-mail address: martin.lotze@uni-greifswald.de (M. Lotze).

Based on these interactions, motor training of hand and finger performance might well profit from somatosensory priming. We therefore explored the efficacy of a combination of rES of the fingertips before 2 weeks of motor training (arm ability training (AAT) [16]) of the left arm in healthy right handed participants. With the current study, we intended to investigate changes of neural representation after rES-primed motor training within the sensorimotor system. AAT is a comprehensive and repetitive motor training that has originally been developed for stroke patients.

In previous studies, we have demonstrated that AAT is capable of increasing motor performance of the non-dominant hand in strongly right-handed healthy participants [17]. In addition, we showed that both tactile resolution and motor performance increased during rES-primed AAT [18]. In the current study, we balanced two groups with and without rES before AAT for performance (AAT scores before training) and demographic data (age and gender) in order to quantify possible additional effects of rES priming.

Using fMRI, we aimed to investigate whether neural substrates underlying performance gain for the rES-primed training were different from those already reported for AAT training alone [17]. We therefore evaluated changes in fMRI activation over training in regions of interest (ROI) preselected on the basis of other studies on long-term motor training [19,20] to evaluate the impact of somatosensory stimulation priming on motor learning. We hypothesized that training should result in an activation decrease of cortical (secondary motor and somatosensory representation) areas, and an activation increase in subcortical (anterior cerebellar hemisphere, basal ganglia) areas.

Materials and methods

Participants

For the rES-primed arm ability training we included 15 healthy, right-handed participants aged 22–28 [25 (mean) \pm 2.2 years (standard deviation); 7 women]. Handedness was determined using the Edinburgh Handedness Inventory with the laterality quotient (LQ), indicating strong dexterity (mean = 98.6 \pm 3.7, range: 89–100). For the non-primed training we recruited 15 participants (24 \pm 3.7 years; 6 women) who were also all strongly right-handed (LQ: 93.5 \pm 5.5; range: 88–100). Both groups were balanced for comparable AAT performance at the start of training.

None of the participants suffered from any neurological disorder or vascular disease (screening by questionnaire), nor were they on any regular medication (contraceptives excluded). Participants were recruited via notice boards at the university campus. Any previous or current regular activity in playing musical instruments was considered an exclusion criterion for study participation. All participants gave their written and informed consent according to the Declaration of Helsinki, and the study was approved by the ethics committee of the Medical Faculty of the University of Greifswald (BB 126/11).

Experimental schedule

The training period extended over two weeks and comprised ten days of arm training. On days 1, 2 and 5, training took place in the laboratory in the presence of the instructor. On all other days, the participants practiced at home. A custom-made software was used (for details, see below) to document the performance times of each task during the training sessions, including graphical feedback to check for plausibility. Motor performance was assessed immediately prior to the first training session and after the second MRI scan. Whenever training or assessment was conducted, the participants were instructed to perform as fast as they could while keeping the

number of errors low, unless the task design included externally paced movements (fist clenching in the scanning session with 1 Hz visually paced).

Training method

Both the rES-group and the non-priming group were enrolled in the active upper limb training. We used a comprehensive finger-hand training developed for stroke patients with moderate upper limb motor impairment (arm ability training; AAT [16]). The AAT targets different sensorimotor abilities such as aiming (i.e. ability to perform quick goal-orientated movements: aiming), arm-hand steadiness (i.e. ability to keep the hand or arm steady; labyrinth, aiming and other trials), wrist-finger speed (i.e. ability to make fast isolated alternating movements of wrist and fingers: tapping), finger dexterity (i.e. ability to manipulate small objects: turning coins; small objects), manual dexterity (i.e. ability to grip and manipulate large objects with hands and arms: bolts and nuts; heavy objects) and visuomotor tracking (i.e. ability to move precisely under continuous visual control: crossing circles; labyrinth) [17,18].

Participants received detailed instructions on the AAT method and the documentation software that was used (AFT 1.2, Platz, Greifswald; Programming by OLIOID GmbH, Berlin, Germany). The time needed for the execution of each of the eight trained tasks was recorded and fed back graphically. Improved performance was indicated by reduced performance time, while the accuracy demands of the tasks were kept constant. All participants performed the training in the same standardized manner with two runs of task performance in a fixed sequence twice a day. The individual improvement was checked for plausibility by the instructor during the assessment sessions. After completion, the whole set was performed a second time, resulting in a total number of four repetitions per task. The duration of the daily training session was approximately 60 minutes.

rES protocol

The participants of the rES-group were stimulated on their left fingertips for 20 minutes/day before they started motor training. The time between stimulation end and training was always less than 20 minutes. No stimulation was used in the non-rES group. The rES sequence consisted of stimulus trains of 1 s (single pulse-duration: 0.2 ms (square), frequency: 20 Hz) and inter-train intervals of 5 s. The sequence was played back from a digital storage device that triggered a standard two-channel TENS device (SM2-AKS, Pierenkemper, Germany) via a custom-made input-channel. The pulses were transmitted via adhesive surface electrodes (1 cm \times 4 cm, Pierenkemper, Germany) fixed on the first and third finger-segments (cathode proximal; see Fig. 1). Stimulation intensity was adjusted to the twofold sensory threshold separately for median and ulnar nerve innervated fingers, resulting in an average initial stimulation intensity of 10.8 \pm 1.5 mA on d1–d3 and 7.8 \pm 0.9 mA on d4 and d5. The adjustment of stimulation current was set according to a previous study exploring the effect of stimulation intensity [1].

Motor performance testing

The performance gain for the left hand was used as an outcome parameter. As a primary outcome variable, we assessed AAT performance as measured by trained doctoral students before and after AAT training separately for the left and right hands. Evaluation of performance was not blinded. The time needed to complete four repetitions of each of the eight training tasks was assessed (Fig. 1B) and changes were averaged as a percentage gain for the complete AAT. As a secondary outcome variable maximum grip force [bar]

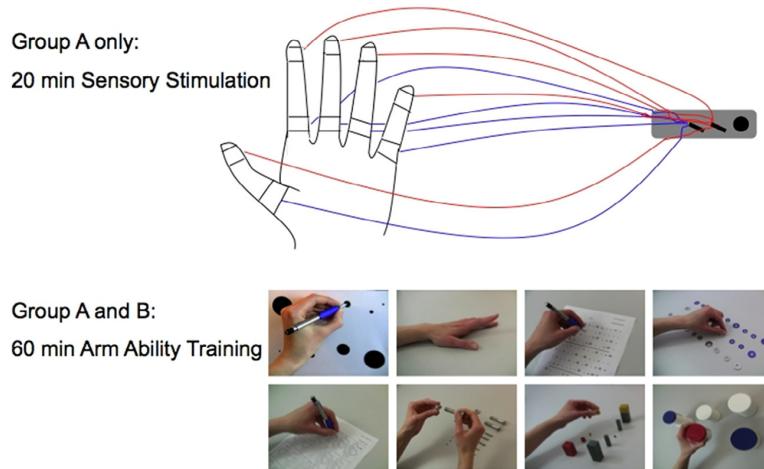


Figure 1. Top (A): The rES-primed participant group was treated with a 20 minute repetitive unattended electric stimulation of the fingertips before one hour of arm ability training. The non-primed group had only arm ability training. Bottom (B): Eight different tasks were applied during the arm ability training (from top left: aiming, tapping, crossing circles, turning coins, labyrinth, nuts and bolts, placing small objects, placing large objects).

was assessed by an experimenter using a vigorimeter (Gebrueder Martin GmbH & Co. KG, Tuttlingen, Germany). Grip force was assessed three times and the average value was calculated before and after training. For "writing", the number of letters copied was counted in 4 blocks, each lasting 20 seconds, and averaged.

Performance gain of the left hand was calculated as percentage changes from the pre-measurement using the following formula: $((\text{post} - \text{pre})/\text{pre}) \times 100$. These were compared between training groups using independent t-tests after testing for relevant difference from normal distribution using the Kolmogorov-Smirnov-Test. We did not use an rmANOVA here because AAT-tasks had to be normalized with percentual improvement before averaging. fMRI-task performance (grip force modulation, finger sequence, writing) was monitored for offline analysis. Repeated measures ANOVAs were conducted with the within-factors group (rES-primed AAT versus non-primed AAT), time (pre, post) and hand (right, left).

fMRI experimental design

MRI-scanning was performed 3–4 days prior to the first day of motor training (pre measurement) and one day after the last block of training had been completed (post measurement). As participants lay on the patient table inside the scanner in a supine position, brain activity was measured during the performance of three different motor tasks with each hand. Two of the tasks were comparable to the trained movements of the AAT, containing the key elements of visually guided movements and repetitive single-digit tapping. The third task was a target force fist-clenching condition for evaluation of transfer effects. Each task was practiced for two minutes prior to scanning in order to achieve a stable performance. A block design was used for each task, alternating five blocks of rest with four blocks of activity. Each block lasted 20 seconds and visual cues indicated either activity (green screen) or rest (blue screen). These cues were transmitted via video projections and a mirror, using Presentation software (version 13.0; Neurobehavioral Systems; Albany, NY, USA 2009), which was triggered by the scanner. Using a pseudorandomized order we assessed each of the following tasks.

Grip force modulation task

A pneumatic rubber ball was squeezed with 1/3 of the maximum grip strength at 1 Hz frequency; visual feedback indicated force am-

plitude as well as a signal for pacing. Both amplitude and frequency of fist clenching were monitored and recorded using a Varioport system that converted pressure levels of the rubber ball into electric signals. Prior to each scanning session the maximum grip strength of either hand was assessed. The participant was asked to squeeze the rubber ball in a maximum voluntary contraction 8–10 times. The participant was then trained for 2 minutes to reach the target force (1/3 of maximum) and frequency (1 Hz).

Tapping of a finger sequence

Twelve numbers were presented visually, and corresponding buttons on a keypad (four-finger-keypads by LUMItouch, Harvard, USA, adapted for each hand) were pressed at 1 Hz frequency. The numbers 2, 3, 4 and 5 corresponded to index, middle, ring and little finger, respectively. An optic fiber system transferred information on key presses to a computer, where it was recorded by Presentation software. The finger sequence was trained outside the scanner room prior to each scanning session.

Writing

The participants were instructed to copy 12 single terms that were arranged in two columns on a horizontally oriented sheet of paper (210 × 297 mm) using a pencil and cursive handwriting. The participants were asked to begin with the column next to the currently writing hand. A line underneath each term provided space for writing. Four sheets of paper with different terms but the same number of letters were used in a pseudorandomized order. All participants of both groups copied the same letters. The sheets were placed on a small desk with an angled board, positioned above the participant's abdomen. Small sandbags supported the upper arm to avoid additional movement, and a double-mirror attached to the head coil allowed for visualization during writing. Between blocks, an assistant standing next to the scanner changed the paper sheets. Performance measure was the number of letters copied, averaged over the four trials.

fMRI measurements

Data acquisition was performed with a Siemens Magnetom Verio 3T-scanner (Siemens; Erlangen, Germany), using a 32-channel head-coil. Field homogeneity was optimized prior to each session using

a shimming sequence. For anatomical images, 176 T1-weighted slices in sagittal orientation were acquired (magnetization prepared rapid gradient echo (MPRAGE); TR = 1.69 s; TE = 2.52 ms; voxel size = $1 \times 1 \times 1$ mm 3 , two times GRAPPA acceleration). Functional images were gathered during motor task performance using T2*-weighted echo planar imaging (EPI) in transversal orientation, parallel to the AC-PC-line (TR = 2.00 s; TE = 30 ms, flip angle = 90°; FoV 192 \times 192 mm 2 ; matrix size = 64 \times 64, voxel size = 3 \times 3 \times 3 mm 3). In total, 90 volumes were obtained, consisting of 34 slices each (thickness of 3 mm, with a 1 mm gap in between slices). To allow for T1 equilibration effects, the first two volumes of each session were discarded. 34 phase and magnitude images were acquired in the same FoV by a gradient echo (flip angle 60°; FoV 192 \times 192 mm 2 ; slice thickness 3 mm; TR = 488 ms; TE₁ = 4.92 ms; TE₂ = 7.38 ms) to calculate a field map in order to correct geometric distortions in EPI images (unwarping [13]).

fMRI data evaluation

Data were analyzed using SPM8 (The Wellcome Trust Centre for Neuroimaging, London, UK) running on Matlab version 7.4 (The MathWorks, Inc.; Natick, MA, USA). The FieldMap toolbox was used to unwarped EPIs that were geometrically distorted due to magnetic field inhomogeneities [21]. To correct for movement artifacts, scans were realigned onto the first scan of each series. The EPIs were then coregistered to the T1-weighted anatomical image and resliced at 3 \times 3 \times 3 mm 3 . The T1-image was segmented and normalized to the Montreal Neurological Institute (MNI) image. To increase the signal-to-noise ratio, smoothing was performed using a 9 \times 9 \times 9 mm 3 full width half maximum (FWHM) Gaussian Kernel filter. Using the general linear model (GLM) we evaluated statistical maps of the main conditions and the comparisons between pre and post measurement for each individual. To perform group analysis, corresponding contrast images were compared in a full-factorial GLM random effects analysis. Within-subjects factors were 'session' (pre and post) and 'task' (grip force modulation, finger sequence, writing).

In a regions-of-interest approach (ROIs) we analyzed the following areas (corrected for multiple comparisons within ROIs, p < 0.05, FWE-corrected; additional cluster threshold: >5 voxel): primary motor cortex (M1), primary somatosensory cortex (S1), secondary motor cortex (supplementary motor area (SMA), premotor cortex (PMC), superior parietal lobe (sensorimotor integration), basal ganglia (putamen, pallidum, caudate), and anterior cerebellar hemisphere (feedforward loops; Larsell lobule IV–VII). Within BA 6 the border between SMA and PMC was defined at the superior frontal sulcus of the MNI-template ($-30 < x < 30$) marking z = 50 as the inferior border of the PMC. Significant brain areas were spatially assigned using SPM Anatomy Toolbox Version 1.7 [22] and, if areas were not defined by Anatomy, using Automated Anatomic Labeling [23]. ROI for S1 was restricted to the finger area (for somatotopic range of the S1 mask see Reference [24]). The S1 mask has also been applied in previous investigations on S1 representations of the fingertips [25].

In order to explore associations of changes in neural representation with changes in motor performance of the trained task (arm ability training), we calculated a linear regression across all 30 participants restricted on the ROIs. We expected specific fMRI-associations with performance gain in the ROIs contralateral to the trained hand.

Results

Group homogeneity at training onset

Participant groups were comparable with respect to age (non-rES: 23.53 \pm 3.70 years, rES: 24.87 \pm 2.23 years; n.s.) and left hand

AAT-performance (aiming: non-rES: 71.69 \pm 6.72 s, rES: 80.39 \pm 10.10 s (n.s.); tapping: non-rES: 65.55 \pm 8.62 s, rES: 70.22 \pm 15.05 s (n.s.); crossing circles: non-rES: 68.40 \pm 16.50 s, rES: 61.10 \pm 12.51 s (n.s.); turning coins: non-rES: 50.74 \pm 7.30 s, rES: 51.60 \pm 9.51 s (n.s.); labyrinth: non-rES: 54.65 \pm 9.41 s, rES: 50.27 \pm 7.86 s (n.s.); bolts and nuts: non-rES: 49.21 \pm 8.78 s, rES: 61.89 \pm 11.64 s (n.s.); small objects: non-rES: 49.59 \pm 11.18 s, rES: 50.97 \pm 7.42 s (n.s.); heavy objects: non-rES: 59.49 \pm 6.67 s, rES: 62.21 \pm 9.52 s (n.s.)) at training onset. However, both training groups differed with respect to their handedness score (Oldfield handedness inventory: non-rES: 93.53 \pm 5.50, rES: 98.6 \pm 3.7; t(28) = 2.96; p < 0.01).

Performance gain during training

Primary outcome variable: For the non-primed group, motor performance averaged across all trained AAT tasks improved by 29.5 \pm 3.5% for the trained left hand. AAT-tasks for the non-trained right hand of this group improved by 20.7 \pm 5.1%. For the rES-primed group, motor performance averaged across all trained tasks improved by 32.9 \pm 5.1% for the trained left hand. AAT-tasks for the non-trained right hand of this group improved by 22.0 \pm 4.4%. For the trained left hand, the effect of rES-primed AAT was larger than those of the non-rES group (averaged AAT-tasks; t(28) = 2.11; Cohen's delta: 0.77; p = 0.044; two-sided; see Fig. 2) and was predominantly driven by finger tapping velocity increase (Cohen's delta: 0.85; the other seven tasks improved with an average effect size of Cohen's delta: 0.27).

As for the secondary, non-trained outcome variable, maximal grip strength for the trained left hand of the non-primed group increased by 1.9%, but by 10.0% for the rES-primed group (t(28) = 2.02; p = 0.027; one-tailed). Writing with the left hand showed a performance increase of 11.0% for the non-primed group, but an 18.6% increase for the rES-primed group (Fig. 3). The difference between training groups was not significant for left hand writing performance (t(28) = 1.45; n.s.), although it showed a moderate effect size (Cohen's delta: 0.53).

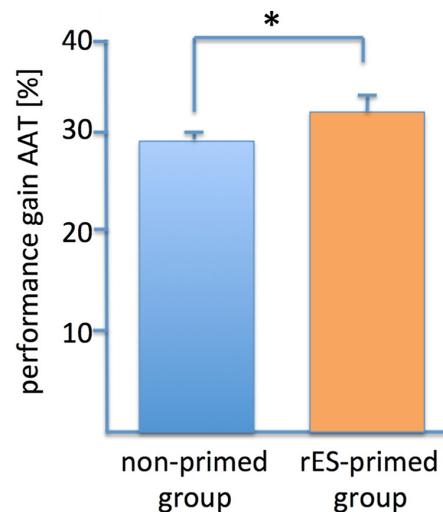


Figure 2. Primary outcome variable: The increase in the primary performance outcome (averaged the arm ability training (AAT) score) in the rES-group (orange bar; n = 15) for the trained left hand was higher (* p < 0.05) than that of the non-primed group (blue bar; n = 15; lines indicate standard error). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

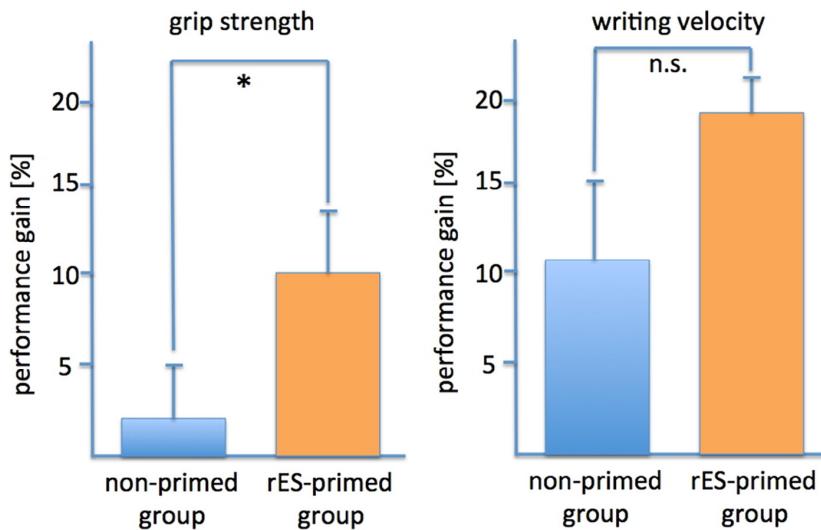


Figure 3. Secondary outcome variables. Left: For grip strength with the left hand the increase in performance in the rES-group was slightly higher (* $p < 0.05$, one-tailed) compared to the non-primed group. Right: For writing with the left hand, the difference between training groups did not reach significance. Lines indicate standard error.

Performance control in the MRI

Repeated-measures ANOVAs did not reveal significant differences between groups for the frequency or amplitude of grip force modulation, finger sequence errors, or number of written words. For the number of written letters, rmANOVA showed a significant effect for both time ($F(14,1) = 37.73$; $p < 0.001$) and hand ($F(14,1) = 180.85$; $p < 0.001$). The number of written letters improved for the trained left hand (rES-primed group: $t(14) = 6.93$; $p \leq 0.001$).

fMRI results

Decrease of fMRI-activation after training (pre minus post)

For the rES-primed group a relevant economization of activation in ROIs was observed only during the finger sequence task with the trained left hand. Significant ROIs comprised bilateral primary sensorimotor cortex, SMA, and superior parietal lobe (see Table 1).

These results were not significantly different from those observed for the non-primed AAT group before [17]. Here left finger sequence performance showed an activation decrease in the dorsolateral prefrontal cortex, ipsilateral S1, ipsilateral parietal lobe, and bilateral SMA. All other tasks performed with either left or right hand showed no significant change over time.

Increase of fMRI-activation after training (post minus pre)

For the rES-primed group a relevant increase of MRI-activation was observed for the grip force modulation task performed with the trained left hand, consisting of an increase of activation in the ipsilateral putamen and pallidum (Table 2). This was a comparable effect as those reported for the non-primed AAT group before [17]. For the writing task with the left hand, an increased activation after training was observed in the right putamen and left anterior cerebellum (Table 2, Fig. 4). Except for the putamen effect, again, the results were the same as those reported for the non-primed AAT group. The finger sequence task showed only an

Table 1
Pre minus post (economization of fMRI-activation).

Task	Area	T-value	p(FWE)	Cluster	x	y	z
Sequence left	M1S1 left	3.86	0.034	19	-12	-36	69
		3.77	0.045	32	-39	-27	42
	M1S1 right	3.82	0.039	77	48	9	33
		3.77	0.044		48	6	42
	SMA	4.52	0.002	90	6	24	45
		3.98	0.014		6	21	60
	Superior parietal left	3.71	0.04	11	-42	-45	57

M1S1: primary sensorimotor cortex; SMA: supplementary motor area.

Table 2
Post minus pre (increase of fMRI-activation).

Task	Area	T-value	p(FWE)	Cluster	x	y	z
Fist clenching left	Putamen left (le)	4.60	0.001	59	-27	0	3
	Pallidum le	4.33	0.001	22	-24	3	0
Writing left	Ant. cerebellum le Larsell lobule 4–7	3.78	0.037	19	-9	-69	-24
	Putamen right (ri)	3.69	0.023	6	27	-3	12
Sequence right	Caudate nucleus ri	3.67	0.026	16	15	15	6
	Putamen right ri	3.52	0.039	17	18	15	3

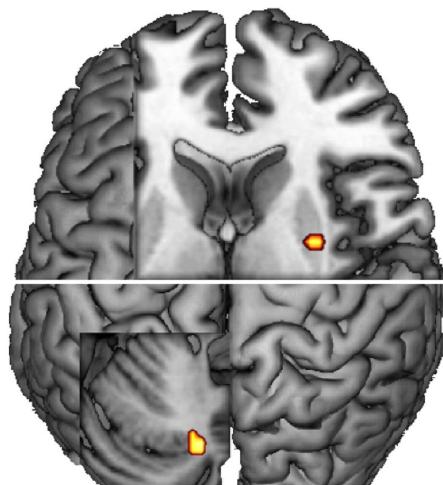


Figure 4. Increase in functional activation in the right putamen and left anterior cerebellum in the rES-primed group for the task “writing with left (trained) hand”. Group analysis for 15 participants with rES-primed AAT over two weeks; post-measurement minus pre-measurement; $p < 0.05$; FWE-corrected for a region of interest analysis.

economization of fMRI-activation for the trained hand. However, finger sequence performed with the non-trained right hand showed increased right caudate and putamen activation (Table 2). For the non-primed group, the differences post minus pre for the finger sequence were centered on the ipsilateral anterior cerebellar hemisphere, the cerebellar vermis and the SPL (BA 5).

Direct comparisons of fMRI-activation maps between groups with and without rES

There were no significant differences between both training groups when comparing changes of functional representation for the trained left hand over time (pre minus post or post minus pre, all three conditions).

Associations between behavioral gain and fMRI activation magnitude

After obtaining negative results from group comparisons, we asked what neural resources drive the performance gain in the AAT task. We hypothesized that an increased activation of the contralateral (right) primary somatosensory cortex (S1) is associated with improved motor performance [26]. We calculated the association of the increase in fMRI-activation for the grip task over time with changes in behavior (AAT-performance increase) and used data of both training groups. Linear regression revealed a positive association of fMRI-activation increase in the right S1 and the performance increase across all AAT-tasks ($t = 3.85$; $p_{\text{FWE}} = 0.039$; MNI-coordinates: 21, -33, 48; see Fig. 5).

Discussion

Repetitive electric stimulation of the fingertips applied before daily arm ability training (AAT) increased performance gain for the trained tasks in the left upper limb. In addition, training effects were also observed for tasks not explicitly trained, indicating transfer and generalization of motor learning. This transfer was substantially larger in the rES group as compared to the non-primed group, indicating a beneficial effect of priming. In the rES group, fMRI showed an increase of basal ganglia and anterior cerebellar representation after training. Performance increase across all trained tasks for all participants (rES-primed and non-primed groups) was associated with an increase of contralateral S1 activation.

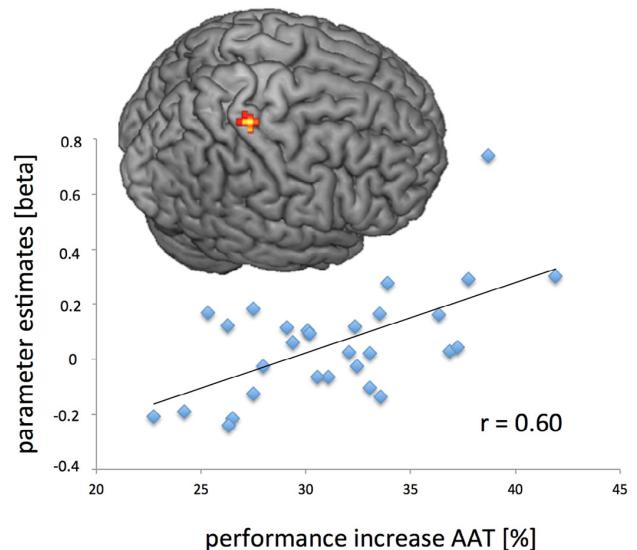


Figure 5. Linear regression analysis between fMRI activation magnitude [beta] increase after training in the contralateral S1 finger area ($t = 3.85$; MNI-coordinates: 21, -33, 48; projection on the rendered MNI-reference brain on top) during grip force modulation and performance increase of the AAT task for both training groups combined. Bottom: plot of linear regression and the corresponding correlation coefficient r .

Increase of performance gain by rES priming

The arm ability training (AAT) is a comprehensive training, which is capable of increasing non-dominant hand motor performance in healthy participants [17]. Our study demonstrates that this performance gain can be further enhanced by rES priming of the fingertips. In addition, the healthy young individuals in our study profited not only for the trained tasks, but also even more for non-trained maximal grip strength of the left hand. In contrast, maximal grip strength was unchanged for non-primed AAT (increase of only 1.9%). This indicates that rES priming might be particularly efficient in driving generalization effects of motor training. It is also conceivable that ceiling effects limited a further improvement in the trained AAT tasks.

It has been demonstrated before that writing with the left or the right hand recruits common neural substrates in the dominant hemisphere [27,28]. Therefore it could be expected that rES priming of the left hand might not relevantly affect motor training of this task, since it should affect excitability of the right hemisphere [29]. However, we observed a quite remarkable effect size for improved velocity of left hand writing over time suggesting that in participants with lower initial performance or in patients with brain damage, rES may lead to significantly increased training outcomes.

The effect of rES amounted to about 3% in addition to the AAT effect in our healthy young group. The effect size observed here (between 0.77 and 0.53) is comparable to strategies increasing cortical excitability in the primary motor cortex directly using anodal TDCS (about 0.59 [30]). This suggests that unattended rES of the fingertips might be an alternative to the more often used anodal TDCS-priming.

Possible mechanism of rES priming

Repeated electrical stimulation of the fingers has been described to result in an increased cortical excitability of the somatosensory cortex as measured by paired pulse median nerve

evoked somatosensory potentials [29], increased fMRI-activation [31,32], parallel to an increase in spatial tactile acuity [6–8]. Typically, the individual gain in discrimination performance was positively correlated with an increase in cortical excitability [29], BOLD signal [31], or EEG-based dipole changes [33].

Somatosensory function and motor function are tightly linked together. Increasing S1-excitability has a direct effect on primary motor cortex excitability (for a recent review see Reference 34). Somatosensory deficits after stroke impair the recovery of voluntary movements [35]. It has been suggested that top-down control of the premotor cortex affects primary somatosensory processing and that the premotor cortex in rodents directly activates layer 5 dendrites in S1 in the absence of temporal coincidence with a bottom-up input [15]. In addition, higher BOLD amplitudes and synchronicity at rest, as measures of cortical activity and synchronicity, are related to better tactile discrimination abilities of the contralateral hand [36]. In humans, increased functional connectivity between SI and MI has been observed following rES [37]. Moreover, an increase in motor excitability after repetitive tactile training has been demonstrated [38].

Accordingly, there are ongoing direct interactions between motor and somatosensory cortex that might be crucial for mediating the beneficial effects of sensory priming of motor behavior. We therefore suggest that an increase in S1 excitability results in a widespread facilitation of primary sensorimotor plasticity and of training effects on both somatosensory and motor performances. This increase in somatosensory performance might especially improve finger tapping rates, an ability that showed the highest effect size of all AAT tasks in the rES-primed group.

Overall changes in functional activation after rES-primed repetitive motor training

We found an economization of ipsi- and contralateral primary and secondary sensorimotor areas (M1, S1, SMA, superior parietal cortex) after training for the finger sequence task. For long term motor training, an economization of cortical representation sites has been previously described especially for motor sequence training [39,40]. Furthermore, expert instrumentalists, in comparison to non-musicians [41] or amateurs [42], show decreased motor activations within the SMA, the PMC, and the ipsilateral M1 during movement performances of varying complexities. Increased activity of the putamen and the ipsilateral cerebellum was found during fist clenching and in the contralateral putamen during writing with the left hand after rES-primed AAT. This finding supports data on changes in functional motor representation after long term training [40]. The cerebellar activations included both anterior and posterior regions, and also anterior parts of the vermis. For the latter, we assume an involvement in the generation of a rhythmic writing component [43].

Limitations of the study

We were not able to observe relevant differences between the rES-primed and the non-primed AAT groups with regard to changes in fMRI-representation of the motor tasks tested. This might be caused by a lack of statistical power, but also by the fact that the fMRI tasks tested were not completely identical to the tasks trained. Changes due to different training procedures might therefore remain unrecognized for the tested conditions. In addition, long-term training procedures show less prominent changes than those present shortly after short-term training [26,40], which might well decrease the effect between training groups, too. In addition we followed the rES protocol developed by the group of Schlieper and Dinse [1]. There might well be developments from other groups

which might be even more advantageous for modulating sensorimotor interaction and for priming upper limb motor training as suggested by other groups [38,44]. Furthermore, the lack of a non-trained control group excludes further conclusions differentiating between habituation and training effects. It is also well possible that the effects of the AAT protocol when applied in healthy young adults show severe ceiling effects, thereby masking possible differences evoked by the addition of priming. An additional limitation is the lack of blinding during data analysis. Blinding should be added in a future study comparing additional effects of somatosensory priming on active motor training.

Conclusions

Repetitive electrical stimulation of the fingertips might be a useful strategy to further enhance motor training gain induced by active motor training. The average improvement of only 3% for the trained tasks makes it possible that the small effects in young healthy adults are due to ceiling. Testing elderly participants or patients is therefore necessary to obtain more information about the beneficial role of rES-based priming. Given the finding of associated primary somatosensory cortex activation increase over all trained participants with training gain, the upper limb motor training should be more focused on somatosensory aspects.

Acknowledgements

We would like to thank Andrea Daniela Walz and Karla Doppl for measurement and data evaluation of the non-primed group. We would also like to thank Evangelia Kaza for help on the development of fMRI-data evaluation scripting, and Nicola Neumann for helpful comments on the manuscript. The study was partially supported by a grant for Martin Lotze from the DFG (LO-795-1).

References

- [1] Schlieper S, Dinse HR. Perceptual improvement following repetitive sensory stimulation depends monotonically on stimulation intensity. *Brain Stimul* 2012;5(4):647–51.
- [2] Dinse HR, Kleibel N, Kalisch T, Ragert P, Wilimzig C, Tegenthoff M. Tactile coactivation resets age-related decline of human tactile discrimination. *Ann Neurol* 2006;60:88–94.
- [3] Kalisch T, Tegenthoff M, Dinse HR. Repetitive electric stimulation elicits enduring improvement of sensorimotor performance in seniors. *Neural Plast* 2010;690531.
- [4] Kattenstroth J-C, Kalisch T, Peters S, Tegenthoff M, Dinse HR. Long-term sensory stimulation therapy improves hand function and restores cortical responsiveness in patients with chronic cerebral lesions. Three single case studies. *Front Hum Neurosci* 2012;6:224.
- [5] David M, Dinse HR, Mainka T, Tegenthoff M, Maier C. High-Frequency repetitive sensory stimulation as intervention to improve sensory loss in Patients with complex regional Pain syndrome I. *Front Neurol* 2015;6:242.
- [6] Sorinola IO, Bateman RW, Mamy K. Effect of somatosensory stimulation of two and three nerves on upper limb function in healthy individuals. *Physiother Res Int* 2012;17(2):74–9.
- [7] Veldman MP, Zijlwendt I, Solnik S, Maffuletti NA, Berghuis KM, Javet M, et al. Direct and crossed effects of somatosensory electrical stimulation on motor learning and neuronal plasticity in humans. *Eur J Appl Physiol* 2015;115(12):2505–19.
- [8] Kowalewski R, Kattenstroth JC, Kalisch T, Dinse HR. Improved acuity and dexterity but unchanged touch and pain thresholds following repetitive sensory stimulation of the fingers. *Neural Plast* 2012;2012:974504.
- [9] Jones EG, Coulter JD, Hendry SH. Intracortical connectivity of architectonic fields in the somatic sensory, motor and parietal cortex of monkeys. *J Comp Neurol* 1978;181:291–347.
- [10] Stepniewska I, Preuss TM, Kaas JH. Architectonics, somatotopic organization, and ipsilateral cortical connections of the primary motor area (M1) of owl monkeys. *J Comp Neurol* 1993;330(2):238–71.
- [11] Wu CW, Kaas JH. Somatosensory cortex of prosimian Galagos: physiological recording, cytoarchitecture, and corticocortical connections of anterior parietal cortex and cortex of the lateral sulcus. *J Comp Neurol* 2003;457(3):263–92.

- [12] Ridding MC, McKay DR, Thompson PD, Miles TS. Changes in corticomotor representations induced by prolonged peripheral nerve stimulation in humans. *Clin Neurophysiol* 2001;112(8):1461–9.
- [13] Kobayashi M, Ng J, Théoret H, Pascual-Leone A. Modulation of intracortical neuronal circuits in human hand motor area by digit stimulation. *Exp Brain Res* 2003;149(1):1–8.
- [14] Classen J, Steinfelder B, Liepert J, Stefan K, Celnik P, Cohen LG, et al. Cutaneomotor integration in humans is somatotopically organized at various levels of the nervous system and is task dependent. *Exp Brain Res* 2000;130(1):48–59.
- [15] Manita S, Suzuki T, Homma C, Matsumoto T, Odagawa M, Yamada K, et al. A top-down cortical circuit for accurate sensory perception. *Neuron* 2015;86(5):1304–16.
- [16] Platz T, Winter T, Müller N, Pinkowski C, Eickhoff C, Mauritz KH. Arm ability training for stroke and traumatic brain injury patients with mild arm paresis: a single-blind, randomized, controlled trial. *Arch Phys Med Rehabil* 2001;82:961–8.
- [17] Walz A, Doppl K, Kaza E, Roschka S, Platz T, Lotze M. Changes in cortical, cerebellar and basal ganglia representation after comprehensive long term unilateral hand motor training. *Behav Brain Res* 2015;278C:393–403.
- [18] Ladda AM, Pfannmöller JP, Kalisch T, Roschka S, Platz T, Dinse HR, et al. Effects of combining 2 weeks of passive sensory stimulation with active hand motor training in healthy adults. *PLoS ONE* 2014;9(1):e84402.
- [19] Doyon J, Penhune V, Ungerleider LG. Distinct contribution of the cortico-striatal and cortico-cerebellar systems to motor skill learning. *Neuropsychologia* 2003;41:252–62.
- [20] Hardwick RM, Rottschy C, Miall RC, Eickhoff SB. A quantitative meta-analysis and review of motor learning in the human brain. *Neuroimage* 2013;67:283–97.
- [21] Hutton C, Bork A, Josephs O, Deichmann R, Ashburner J, Turner R. Image distortion correction in fMRI: a quantitative evaluation. *Neuroimage* 2002;16:217–40.
- [22] Eickhoff SB, Stephan KE, Mohlberg H, Grefkes C, Fink GR, Amunts K, et al. A new SPM toolbox for combining probabilistic cytoarchitectonic maps and functional imaging data. *Neuroimage* 2005;25:1325–35.
- [23] Tzourio-Mazoyer N, Landau B, Papathanassiou D, Crivello F, Etard O, Delcroix N, et al. Automated anatomical labeling of activations in SPM using a macroscopic anatomical parcellation of the MNI MRI single-subject brain. *Neuroimage* 2002;15:273–89.
- [24] Weibull A, Björkman A, Hall H, Rosen B, Lundborg G, Svensson J. Optimizing the mapping of finger areas in primary somatosensory cortex using functional MRI. *Magn Reson Imaging* 2008;26(10):1342–51.
- [25] Pfannmöller JP, Schweitzer R, Lotze M. An automated analysis protocol for high resolution BOLD-fMRI mapping of the fingertip somatotopy in Brodmann area 3b. *Magn Reson Imaging* 2015;43(2):479–86.
- [26] Lotze M, Braun C, Birbaumer N, Anders S, Cohen LG. Motor learning elicited by voluntary drive. *Brain* 2003;126(4):866–72.
- [27] Rijntjes M, Dettmers C, Büchel C, Kiebel S, Frackowiak RS, Weiller C. A blueprint for movement: functional and anatomical representations in the human motor system. *J Neurosci* 1999;19(18):8043–8.
- [28] Planton S, Jucla M, Roux F-E, De Monet J-F. The “handwriting brain”: a meta-analysis of neuroimaging studies of motor versus orthographic processes. *Cortex* 2013;49:2772–87.
- [29] Höffken O, Veit M, Knossalla F, Lissek S, Blüm B, Ragert P, et al. Sustained increase of somatosensory cortex excitability by tactile coactivation studied by paired median nerve stimulation in humans correlates with perceptual gain. *J Physiol* 2007;584(Pt 2):463–71.
- [30] Kang N, Summers JJ, Cauraugh JH. Transcranial direct current stimulation facilitates motor learning post-stroke: a systematic review and meta-analysis. *J Neurol Neurosurg Psychiatry* 2016;87(4):345–55.
- [31] Pieger B, Foerster AF, Ragert P, Dinse HR, Schwenkreis P, Malin JP, et al. Functional imaging of perceptual learning in human primary and secondary somatosensory cortex. *Neuron* 2003;40(3):643–53.
- [32] Hodzic A, Veit R, Karim AA, Erb M, Godde B. Improvement and decline in tactile discrimination behavior after cortical plasticity induced by passive tactile coactivation. *J Neurosci* 2004;24(2):442–6.
- [33] Pieger B, Dinse HR, Ragert P, Schwenkreis P, Malin JP, Tegenthoff M. Shifts in cortical representations predict human discrimination improvement. *Proc Natl Acad Sci USA* 2001;98(21):12255–60.
- [34] Veldman MP, Maffuletti NA, Hallett M, Zijdewind I, Hortobágyi T. Direct and crossed effects of somatosensory stimulation on neuronal excitability and motor performance in humans. *Neurosci Biobehav Rev* 2014;47:22–35.
- [35] Nudo RJ, Friel KM, Delia SW. Role of sensory deficits in motor impairments after injury to primary motor cortex. *Neuropharmacology* 2000;39(5):733–42.
- [36] Haag LM, Heba S, Lenz M, Glaubitz B, Höffken O, Kalisch T, et al. Resting BOLD fluctuations in the primary somatosensory cortex correlate with tactile acuity. *Cortex* 2015;64:20–8.
- [37] Freyer F, Reinacher M, Nolte G, Dinse HR, Ritter P. Repetitive tactile stimulation changes resting-state functional connectivity—implications for treatment of sensorimotor decline. *Front Hum Neurosci* 2012;6:144.
- [38] Schabrun SM, Ridding MC, Galea MP, Hodges PW, Chipchase LS. Primary sensory and motor cortex excitability are co-modulated in response to peripheral electrical nerve stimulation. *PLoS ONE* 2012;7(12):e51298.
- [39] Shadmehr R, Krakauer JW. A computational neuroanatomy for motor control. *Exp Brain Res* 2008;185(3):359–81.
- [40] Dayan E, Cohen LG. Neuroplasticity subserving motor skill learning. *Neuron* 2011;72(3):443–54.
- [41] Lotze M, Scheler G, Tan HRM, Braun C, Birbaumer N. The musician's brain: functional imaging of amateurs and professionals during performance and imagery. *Neuroimage* 2003;20:1817–29.
- [42] Pau S, Jahn G, Sakreida K, Domini M, Lotze M. Encoding and recall of finger sequences in experienced pianists compared to musically naives: a combined behavioural and functional imaging study. *Neuroimage* 2013;64:379–87.
- [43] Penhune VB, Zattore RJ, Evans AC. Cerebellar contributions to motor timing: a PET study of auditory and visual rhythm reproduction. *J Cogn Neurosci* 1998;10(6):752–65.
- [44] Chipchase LS, Schabrun SM, Hodges PW. Peripheral electrical stimulation to induce cortical plasticity: a systematic review of stimulus parameters. *Clin Neurophysiol* 2011;122(3):456–63.