Universität Basel

Fakultät für Psychologie

Brain Plasticity Induced by Motor and Cognitive Interventions in Health and Pathology

A cumulative dissertation submitted to the Faculty of Psychology, University of Basel, in partial fulfilment of the requirements for the degree of Doctor of Philosophy by

Priska Zuber

from Basel, Switzerland

Basel, 2021

Universität Basel

Fakultät für Psychologie

Approved by the Faculty of Psychology at the request of

Professor Dr. med. Dominique J.-F. de Quervain

Dr. phil. Stefano Magon

Date of the doctoral examination:

Dean of the Faculty of Psychology

Universität
Basel

Fakultät für
Psychologie

Declaration of Scientific Fairness

I, **Priska Zuber**, hereby declare that the present work was written independently without the help of third parties and without the use of any means other than those indicated. Sources used for help are marked as such. The manuscripts published or submitted for publication in journals were prepared in cooperation with the co-authors and were not published elsewhere by any of the participants, submitted for publication, or submitted to any other examination authority as a qualification paper. These are the following manuscripts:

A) **Zuber, P.,** Tsagkas, C., Papadopoulou, A., Huerbin, M., Gaetano, L., Geiter, E., Altermatt, A., Parmar, K., Ettlin, T., Schuster-Amft, C., Suica, Z., Alrasheed, H., Wuerfel, J., Kesselring, J., Kappos, L., Sprenger, T., Magon, S. (2020). Efficacy of Inpatient Multidisciplinary Rehabilitation in Multiple Sclerosis: Behavioural and fMRI Results. *Journal of Neurology*, 1-10.

B) **Zuber, P.,** Geiter, E., de Quervain, D., Magon, S. (2021). Development of a Model-based Working Memory Training with and without Distractor Inhibition and Investigation of its Comparative Efficacy: A Randomized Study on Healthy Adults. *Frontiers in Aging Neuroscience. (submitted for publication)*

C) **Zuber, P.,** Gaetano, L., Griffa, A., Huerbin, M., Pedullà, L., Bonzano, L., Altermatt, A., Tsagkas, C., Parmar, K., Hagmann, P., Wuerfel, J., Kappos, L., Sprenger, T., Sporns, O., Magon, S. (2021). Additive and Interaction Effects of Working Memory and Motor Sequence Trainings on Brain Functional Connectivity. *Cortex. (submitted for publication)*

Basel, 12. May 2021

Priska Zuber

Abstract

Plasticity, the way the brain can adapt to environmental or intrinsic changes, is the cornerstone of human behavior. Plasticity does not only harbor the possibility to learn or improve new motor or cognitive skills, but also displays the basis for restoring functioning after brain damage. Brain plasticity can thus be induced by various activities and is most prominently investigated following interventions with the aim to enhance cognition in the healthy population or rehabilitative strategies of motor and cognitive functions in pathological conditions. Although it is widely acknowledged that plastic changes are possible after activity-dependent interventions, improvements on untrained functional domains are still not fully comprehended. Furthermore, the previous focus on establishing the efficacy has shifted towards the understanding of behavioral and functional underlying mechanisms of interventions. Three studies presented within the frame of this thesis aimed at investigating mechanisms of brain plasticity in health and pathology induced by rehabilitative, motor and cognitive interventions.

In the first study (Zuber et al., 2020), we examined the efficacy in symptom improvement and underlying brain mechanisms of an inpatient personalized multidisciplinary rehabilitation program in patients with multiple sclerosis using functional Magnetic Resonance Imaging (fMRI). The multidisciplinary rehabilitation led to improved fatigue, walking ability as well as quality of life and a more effective recruitment of cerebellar and prefrontal brain regions in patients with multiple sclerosis.

Two studies in healthy participants aimed at targeting current challenges in the cognitive training research by studying underlying cognitive and motor mechanisms of working memory training. In study B (Zuber, Geiter, de Quervain, & Magon, 2021), we compared a novel model-based working memory training with and without distractor inhibition to a dual-n-back and active control training in order to study distractor inhibition as a task-related process with the potential to render near and far transfer effects in healthy elderly adults. Working memory capacity was improved only following the model-based training with distractor inhibition, suggesting the novel training to be a promising approach in improving working memory in healthy old adults. In study C (Zuber, Gaetano, et al., 2021), we studied the interactive and additive effects of working memory and motor sequence learning training behaviorally and by resting state functional connectivity. Results indicate a relevance of the sequential order of training performance, with increased functional connectivity between a complex network of parietal, temporal and motor brain regions, specifically when motor training was performed before or combined with working memory training.

The results of those three studies indicate plastic changes following rehabilitation in patients with multiple sclerosis and motor and cognitive interventions in healthy people. It can thus be concluded that brain plasticity following interventions in health and pathology is the result of an interplay between various behavioral and functional mechanisms. Hence, this thesis highlights the importance of identifying underlying neural and behavioral processes by theory-driven approaches in methodologically well controlled studies as the basis for translating neuroscientific research into the clinical setting.

Acknowledgements

In the course of my doctorate, I had the honour of being part of a multidisciplinary environment and work together with excellent researchers to whom I would like to express my gratitude to.

From the first thoughts about starting my PhD and throughout the whole process I always felt competently advised, encouraged and most importantly constantly supported by my two supervisors, Prof. Dr. Dominique de Quervain and Dr. Stefano Magon. Specifically, I want to express my deep appreciation to Prof. de Quervain for giving me the opportunity to experience scientific work as a benevolent, supportive collaboration which formed the basis for me to investigate my research questions with great pleasure and motivation. I further want to express my profound gratitude towards Dr. Magon for not only initiating my interest for neuroscientific research, but more so for introducing me to the scientific world with all his expertise and standing by me through the whole time of my PhD. Further thanks go to my external supervisor Prof. Dr. Gianpaolo Basso for offering his expertise and supporting me from afar.

Completing my PhD at the intersection of different disciplines has given me the unique opportunity of working together with wonderful people. I thus want to thank the whole team of the Transfaculty Research Platform Molecular and Cognitive Neurosciences (MCN) of the University of Basel. Specifically, I want to thank M.Sc. Anja Zimmer, the kindest and most supportive co-PhD student and dear friend since the first semester of my studies at the University of Basel. I also want to thank Dr. Bernhard Fehlmann for always providing me with great advice. Furthermore, I want to thank the whole team of the Medical Image Analysis Center (MIAC), led by Dr. Jens Wuerfel, where my research career took its first steps as an assistant to Dr. Magon. Having MIAC as a work environment gave me the possibility to collaborate with computer scientists, physicians, engineers and physicists, which thought me the importance of taking perspective in order to proceed. I specifically appreciated working with B.Sc. Emanuel Geiter, which resulted in a fun and creative smartphone application we jokingly call our “baby”. I also want to thank Dr. Anna Altermatt and Dr. Charidimos Tsagkas for all discussions around research questions, concerns or future plans and for celebrating each little step in our different paths with an afterwork aperitive. I am deeply grateful for the fun and intelligent people I met and now call my dear friends.

I also want to thank all students and research assistants I worked with, that is M.Sc. Jan Bitterli, M.Sc. Valentina Halasi, B.Sc. Gresa Mazreku, B.Sc. Martina Foresti, M.Sc. Lauriane Arnegger, M.Sc. Vera Nina Looser, M.Sc. Elena Pauli, M.Sc. Julian Gygax, M.Sc. Galya Iseli and M.Sc. Melike Sahin. They were the reason that a large and intense project such as study B could be carried out in a fun, easy but also thorough way.

Further thanks apply to the Swiss National Science Foundation (SNSF) for financially supporting my PhD with the research grant Doc.CH: P0BSP1_181741.

Last but not least I would like to express my sincere thanks to the dear people – and my little dog Quincy – around me for being my basis and source of strength allowing for me to grow continuously. I want to thank my parents, my dear brother Andreas as well as Alexia, Josefina and Xenia for always being there for me. Finally, I am endlessly grateful to Giuliano, my beloved partner, for unconditionally supporting and always encouraging me in his sincere and heartfelt way.

Table of Contents

Abbreviations

BOLD	Blood Oxygen Level Dependent
CNS	Central Nervous System
FC	Functional Connectivity
fMRI	functional Magnetic Resonance Imaging
MS	Multiple Sclerosis
MSL	Motor Sequence Learning
WM	Working Memory
WHO	World Health Organisation

"*Swiftly the brain becomes an enchanted loom, where millions of flashing shuttles weave a dissolving pattern - always a meaningful pattern - though never an abiding one; a shifting harmony of sub-patterns*."

-

Charles Sherrington, 1941

1 Introduction

As stated by Santiago Ramon y Cajal in 1928, up until then the adult brain was thought to be an organ where "[…] nerve paths are something fixed, ended, immutable. Everything may die, nothing may be regenerated" (Cajal, 1928). However, after the detection of the function of neurons and synapses – amongst others by Charles Sherrington, Donald Hebb was the first to introduce the concept of neural plasticity as the repeated firing of neurons which can lead to structural changes and wired synapses (Power & Schlaggar, 2017; Von Bernhardi, Eugenín-Von Bernhardi, & Eugenín, 2017). Animal and human research later confirmed that the cortex has the capacity to reorganize and produce neurons up to adult age (Power & Schlaggar, 2017; Raymont & Grafman, 2006; Ward, 2005), leading to the understanding of the plastic brain as we have today.

Nowadays, *neuroplasticity* is defined as "[…] the ability of the nervous system to respond to intrinsic and extrinsic stimuli by reorganizing its structure, function and connections […]" (Cramer et al., 2011) and displays the cornerstone of the normal functioning of the brain as well as neural development (Von Bernhardi et al., 2017). Following this definition, neuroplasticity can be described as adaptive, when leading to positive changes in functioning or as maladaptive, when leading to negative changes (Cramer et al., 2011; Kloos, Gomes-Osman, & Boyd, 2020). Therefore, neuroplasticity can be understood on the cellular level, as a response to the environment, in the context of cognitive functioning, following motor or cognitive learning or as a response to aging or pathological conditions (Cramer et al., 2011; Von Bernhardi et al., 2017; Ward, 2005; Willis & Schaie, 2009). Within this comprehensive understanding of neural plasticity, scientists and clinicians are likewise interested in investigating underlying mechanisms, interventions and translations to promote adaptive neuroplasticity in clinical conditions but also in the healthy population (Kloos et al., 2020).

Although our understanding of the brain's plasticity changed since Cajal and it is widely acknowledged that plastic changes occur following training or rehabilitation, the extent of improvement on various functional domains following such interventions is still not fully comprehended. While the focus of previous interventional studies laid on establishing the efficacy, it has become clear today that their behavioral and functional underlying mechanisms have yet to be fully understood. For this reason, this thesis aimed at investigating brain plasticity in health and pathology induced by rehabilitative, motor and cognitive interventions with focusing on their underlying neural and behavioral mechanisms. Specifically, the first part of the thesis will introduce the current state of research in neuroplasticity in the context of pathology with a focus on multiple sclerosis and its rehabilitation. Further, functional Magnetic Resonance Imaging will be introduced as a tool to investigate neuroplasticity. At this point, the first original research paper **A)** entitled "*Efficacy of inpatient multidisciplinary rehabilitation in multiple sclerosis: behavioral and* *fMRI results"* will be presented. In this study, we investigated the efficacy and underlying brain mechanisms of a personalized inpatient multidisciplinary rehabilitation program in people with multiple sclerosis. The results indicated that multidisciplinary rehabilitation can induce efficient recruitment of brain regions and leads to an improvement in highly impacting symptoms of multiple sclerosis. For this study, my contribution covers the collection and curation of the behavioral and imaging data, their formal analyses and writing of the original manuscript.

The second part of this thesis will then delve deeper into the topic of cognitive and neural plasticity as a result of learning and memory in the healthy young and aging population. The focus will lie on current challenges and possible motor and cognitive mechanisms underlying working memory trainings, presented by two original studies **B)** entitled "*Development of a Model-based Working Memory Training with and without Distractor Inhibition and Investigation of its Comparative Efficacy"* and **C)** "*Additive and interaction effects of working memory and motor sequence trainings on brain functional connectivity"*. Study **B** aimed at investigating the comparative efficacy of a model based working memory training with and without distractor inhibition in improving working memory capacity in healthy old adults. The results showed increased working memory capacity only following the model-based training with distractor inhibition, suggesting it to be a promising approach for training working memory in healthy old adults. My contribution covers the conceptualization of the working memory training and the clinical trial, data collection and curation, formal analyses and writing the original manuscript. In study **C**, we studied the differences in functional connectivity induced by sequential and combined training of working memory and motor sequence learning in healthy young adults. The results indicate distinct patterns of resting state functional connectivity changes which were modulated by the order of training performance. My contribution covers the collection and curation of behavioral and imaging data, formal analyses of the behavioral data as well as writing the original manuscript.

2 Plasticity in the Context of Pathology

Brain injury or neurological diseases can lead to structural, molecular and functional changes in the brain (Kleim, 2011), which can result in a large range of symptoms. Since it has been shown that injury-related damage in the brain induce changes in unrelated brain regions, it has been suggested that those neuroplastic changes can be targeted by rehabilitative interventions which act as a re-learning process with the aim to reduced impairment (Kleim, 2011; Ward, 2005). Indeed, it has been shown that structural and functional plasticity due to sensorimotor learning and brain injury interact and can thus be targeted in order to reorganize during recovery (Nudo, 2003). Hence, restoring and regenerating lost functions is of utmost importance for patients and clinicians (Ward, 2005). However, neuroplastic changes after brain damage are not uniform and can be categorized in four functional neuroplastic mechanisms (Grafman, 2000). First, *homologous area adaptation* describes the compensation of cognitive function through damage in a specific brain region by the corresponding region in the opposite brain hemisphere. Since the shift to the new brain area can lead in overrepresentation of the cognitive function which in turn can impair the learning of new skills that would have originally processed by this brain region, homologous area adaptation is assumed to be more common in children and adolescents still in the developmental phase. Second, *cross-modal reassignment* describes a form of neuroplasticity where sensory inputs are redirected in the brain regions which have experienced deprivation of its main inputs due to damage. However, cross-modal reassignment is limited by the fact that some cells in the brain are highly specialized, which makes a reassignment of other forms of input unlikely. Third, *compensatory masquerade* describes the use of a non-affected cognitive process to perform a task that used to be carried out by the damaged cognitive process. The last form of functional neuroplasticity is *map expansion*, which can be described as the temporary or persistent enlargement of cortical regions resulting from skill training and practice. It is assumed that map expansion can either recruit neurons into the regional network responsible for a function or that the whole network is activated until the exact representational region is selected. The latter would explain that increases but also decreases in brain activation occur following practice (Grafman, 2000). In order to restore functions after brain damage or disease, it has been stated that activity-induced neuroplasticity understood as map expansion displays the interface to recovery and is specifically of interest the first months following the damaging event (Horton, Fahle, Mulder, & Trauzettel-Klosinski, 2017; Nudo, 2003). In the following chapters, functional neuroplasticity as the basis for rehabilitation after brain injury or disease will be discussed in the context of multiple sclerosis.

2.1 Multiple Sclerosis

Multiple sclerosis (MS) is the most common neurodegenerative and inflammatory disease of the central nervous system globally affecting around 2.3 million people with a peak incidence in young adulthood (Kamm, Uitdehaag, & Polman, 2014; Reich, Lucchinetti, & Calabresi, 2018; Thompson, Baranzini, Geurts, Hemmer, & Ciccarelli, 2018). It is well described that MS occurs more often in women, where a 2.5% lifetime risk has been described compared to 1.4% in men (Kamm et al., 2014). Several risk factors have been associated with the development of MS. First, environmental factors such as higher geographic latitude and thus hypothetical vitamin D deficiency, smoking, obesity or mononucleosis as a result of infection with the Epstein-Barr virus have been associated with higher risk of MS (Kamm et al., 2014; Reich et al., 2018). Second, familial frequency and being a carrier of the HLA DRB1*150 allele, which is involved in the development of autoimmune diseases, have been suggested as genetic risk factors for MS (Reich et al., 2018; Thompson et al., 2018). Although the association of those factors with the development of MS has been epidemiologically established, their role regarding the etiology of MS is still poorly understood (Reich et al., 2018).

In MS, central nervous system (CNS) tissue damage arises as a result of an interaction between the immune system, glia and neurons, leading to demyelination, axonal injury and inflammation (Kamm et al., 2014; Reich et al., 2018). The hallmark of this disorder are focal inflammatory events occurring in the CNS, so-called white matter lesions or *plaques*. In the early disease stages, white matter tissue damage is initiated by lymphocytes, mainly T cells, that enter the CNS via the blood-brain barrier, are reactivated by antigen presenting cells, stimulate microglia as well as astrocytes and induce plasma cell antibody production leading to demyelination (Kamm et al., 2014). Following this acute inflammatory processes, the demyelinated plaques can either be remyelinated, stay chronically inactive or result in slow but persisting myelin degeneration in *smoldering* lesions (Reich et al., 2018). MS lesions can be located in the brain white matter, brain stem, spinal cord or the optic nerve, but also at the junction between the white and grey matter or entirely within the grey matter (Reich et al., 2018).

At least partially due to the occurrence of lesions within the whole CNS, MS is characterized by a polysymptomatic clinical picture affecting cognition, mobility, bladder function, hand function, coordination as well as vision and can lead to depression, pain, fatigue, walking disability, spasticity, sensory symptoms and tremor (Kister et al., 2013). It has been described that sensory symptoms, fatigue and cognitive impairment are the most prevalent symptoms reported by patients within the first year of the disease accompanied by motor impairments (Kister et al., 2013). Based on the presence of acutely or sub-acutely occurring clinical worsening – also known as relapses –, as well as evident gradual progression of disability over a period of time, MS can be divided into clinical phenotypes (Lublin et al., 2014). Whereas *primary progressive MS* is characterized by gradual clinical worsening with progressive accumulation of disability from onset, *relapse-onset MS* is the most common disease phenotype and includes relapsing and remitting phases of the disease. In these patients, disease onset is marked by a single episode, so-called clinical isolated syndrome, affecting the optic nerve, brainstem or spinal cord, which can lead to clinical definite MS when it is followed by a second episode or relapse (Thompson et al., 2018). Within relapse-onset MS a further distinction can be made between *relapsing-remitting MS* that is described by relapses alternated with remitting phases and *secondary-progressive MS*, which is characterized by additional worsening of symptoms without remission after initial relapsing course (Lublin et al., 2014; Thompson et al., 2018).

2.2 Plasticity induced by Rehabilitation in Multiple Sclerosis

As outlined in the previous section, various functional domains can be affected in patients due to the polysymptomatic clinical picture of MS. In addition, MS is characterized by unpredictable changes of the pathology during the disease course which leads to inter- and intraindividual variability in the clinical pattern of MS in patients (Beer, Khan, & Kesselring, 2012). Thus, MS has not only been associated with high impact on personal life as well as decreased quality of life in patients, but leads to substantial socio-economic consequences due to the high prevalence of long-term disability as a result of the early onset of the disease (Beer et al., 2012; Khan, Amatya, Galea, Gonzenbach, & Kesselring, 2017).

Although disease-modifying drugs have been described to be effective in reducing inflammation and the number of relapses in MS, they lack the ability to delay progression and disability (Feinstein, Freeman, & Lo, 2015). However, it has been suggested that recovery-based interventions have the possibility to restore brain activity and structures at elevated levels of disability in MS. This has been attributed to the fact that brain activation changes together with recovery of motor functions were observed after decreased inflammation due to spontaneous remission after relapses (Lipp & Tomassini, 2015). Indeed, it has been hypothesized that brain damage, disability and functional reorganization are related in MS (Schoonheim, Geurts, & Barkhof, 2010). Additionally, activity-dependent interventions have been described to enhance recovery after relapses which can reduce the extent of residual disability and increase long-term functioning in people with MS (Rooney, Albalawi, & Paul, 2020). For this reason, interventions that promote activity-dependent neuroplasticity play a crucial role in the symptomatic treatment of MS (Tomassini et al., 2012).

According to the World Health Organization (WHO), rehabilitation is defined as “a set of interventions designed to optimize functioning and reduce disability in individuals with health conditions in interaction with their environment.” (World Health Organization, 2020). Since MS can lead to cumulative effects of impairment and disability in multiple functional domains, rehabilitation in MS is characterized by an interdisciplinary, goal-oriented and individualized care program including different rehabilitative therapy modules (Amatya, Khan, & Galea, 2019; Khan & Amatya, 2017). Rehabilitative interventions in MS include physical therapy programs, cognitive or psychological interventions, occupational therapy, whole-body vibration, hippotherapy, vocational therapy, information provision interventions, symptom-specific but also multidisciplinary rehabilitation programs (Khan & Amatya, 2017). Results of two recent systematic reviews that investigated the efficacy of rehabilitative interventions in MS revealed moderate to strong evidence for the improvement of mobility, muscular strength, fatigue and quality of life following physical and exercise therapy as well as strong evidence for fatigue management programs. Further, moderate evidence was reported for strength training, energy conservation, cognitive behavioral therapy or multidisciplinary rehabilitation and low quality evidence for specific interventions such as exercise therapy for cognition, hippotherapy, neuropsychological or memory rehabilitation, as well as dietary interventions (Amatya et al., 2019; Khan & Amatya, 2017). Following these results it has been concluded that future research should focus on the investigation of interventions that can be translated in personalized multidisciplinary rehabilitation (Amatya et al., 2019).

Indeed, it has been stated that future rehabilitation trials should lay the focus on multidisciplinary rehabilitative approaches and test whether they may efficiently target neural plasticity in MS (Feinstein et al., 2015; Tomassini et al., 2012). Thus, although the possibility of promoting plastic changes by activity-dependent interventions has been described along evidence for the efficacy of rehabilitative strategies, the underlying neural mechanisms of recovery through individualized multidisciplinary rehabilitation in MS needs to be further evaluated. One way of investigating neuroplasticity induced by rehabilitation in an non-invasive way is functional Magnetic Resonance Imaging (De Giglio, Tommasin, Petsas, & Pantano, 2018). The following chapter will give a short overview of functional Magnetic Resonance Imaging as a technique to assess neuroplasticity, followed by the presentation of the original research paper (Zuber et al., 2020) displaying one of the first studies to investigate neuroplastic changes after a personalized multidisciplinary rehabilitation program in people with MS assessed by functional Magnetic Resonance Imaging.

2.3 Functional Magnetic Resonance Imaging to Measure Neuroplasticity

Functional Magnetic Resonance Imaging (fMRI) is an imaging method allowing to investigate the time-varying changes in brain metabolism on the basis of blood flow (Glover, 2011). It allows to investigate the activity of the brain at rest or related to specific tasks and has been suggested to reflect neuronal signaling (Matthews & Jezzard, 2004). The physiological basis of fMRI arises from the increased neural signaling following the activation of a brain region leading to higher demands of energy in the form of substrates for the energy metabolism, which in turn results in an oxygen deficit in the local brain region. In order to restore the local oxygen deficit, the blood flow is increased. As a consequence, activation of the brain region leads to decreased deoxygenated hemoglobin and increased oxygenated hemoglobin, also termed the hemodynamic response of the neural event. The two forms of oxygenation of hemoglobin display different local magnetic fields with deoxygenated hemoglobin being paramagnetic – which suppresses the fMRI signal – and oxygenated hemoglobin being diamagnetic. With decreasing deoxygenated hemoglobin, the fMRI signal increases. These changes in the magnetic field induced by oxygenation concentration are called Blood Oxygen Level Dependent (BOLD) contrast (Glover, 2011).

fMRI experiments can either include the acquisition of BOLD contrast images for a fixed amount of time during a motor or cognitive task in the context of task-related fMRI, or during the brain at rest as so-called resting state fMRI (Chen & Glover, 2015). Task-related fMRI experiments are typically designed as *block designs* with alternating control or experimental conditions that are later contrasted to focus on the cognitive mechanism of interest or as *event related*, where BOLD data is acquired during the discrete presentation of stimuli. In order to identify brain regions activated by task fMRI, statistics are inferred from the BOLD signal variations resulting from the experimental model. In contrary, resting state fMRI hypothesizes spontaneous, synchronized activity fluctuations in distinct regions when the brain is at rest, which have been identified and described as several networks of the resting brain (Chen & Glover, 2015; Damoiseaux et al., 2006; Matthews & Jezzard, 2004).

Before statistical interferences can be drawn, collected time series of both task-related and resting state fMRI require preprocessing, in order to remove confounding factors unrelated to the hemodynamic response. Preprocessing steps include 1) a quality assurance of the acquired data, 2) slice time correction due to inconsistent acquisition of time among brain slices, 3) correction of the participants head motion, 4) distortion correction due to inhomogeneity in the magnetic field, 5) temporal filtering in order to eliminate noisy frequencies, 6) spatial smoothing in order to improve the signal to noise ratio, 7) physiological noise correction of e.g. cardiac pulse or respiration, 8) co-registration of the functional to structural MRI images as well as 9) spatial normalization in order to standardize each participants brain to a template brain (Chen & Glover, 2015). Although preprocessing steps are able to correct for various types of noise that can occur in the BOLD acquisition, the low temporal resolution, signal dropout or spatial distortions are limitations of fMRI. However, the relatively high spatial resolution and availability in various research fields is considered a strength of fMRI (Glover, 2011). Following the preprocessing steps, task-related fMRI data analysis is typically done in a mass-univariate way, where each voxel's signal is modelled independently from the others by applying t-tests, correlations, general linear models or multivariate analyses on either single-subject or group level (Chen & Glover, 2015). Resting state fMRI data analysis focuses on the statistical dependencies between different brain regions, named functional connectivity (FC) (Chen & Glover, 2015). FC can be inferred via seed-based analyses, independent component analysis or graph theory analyses (Smitha et al., 2017).

Due to its strengths, task-related fMRI is a broadly used technique to investigate neuroplasticity (Reid, Boyd, Cunnington, & Rose, 2016). Amongst various task-fMRI paradigms, motor sequence learning (MSL) has been introduced to investigate motor skills after interventions in both patients with MS and healthy adults (Deroost, Smetcoren, Vandenbossche, & Hooghe, 2014; Tacchino et al., 2015). In a MSL task, participants are asked to respond to a series of visual targets, by pressing buttons with their fingers. The instruction on the presence of a sequence can either be implicit or explicit, tapping different learning processes of motor skill learning (Deroost et al., 2014). However, specifically the investigation of neuroplastic changes with task-fMRI following rehabilitation harbors challenges. Specifically, 1) subject variability due to anatomy or location, type and timing of insult, 2) biological ambiguity in activation due to compensatory mechanisms, strategic improvements of task performance, task difficulty or disinhibition, 3) methodological considerations related to pre-processing steps or analyses or 4) disease-related confounds such as acute effects, altered hemodynamic response or head movement can hamper the interpretation of activity patterns in task-related fMRI results following interventions (Reid et al., 2016). Nevertheless, fMRI is widely used in MS to study disease-related changes in brain activation or neuroplastic changes following pharmacological therapy or rehabilitative interventions. Studies are either designed cross-sectional or longitudinal and are typically controlled with a sample of healthy participants (De Giglio et al., 2016). Functional changes following clinical improvements as well as brain regions involved in the recovery were described by fMRI studies in MS, specifically following rehabilitative interventions in brain regions involved in learning processes (De Giglio et al., 2016). However, due to the challenges that can arise in the investigation of neuroplasticity following rehabilitation, longitudinal fMRI studies that describe the functional changes following personalized rehabilitation strategies are scarce (De Giglio et al., 2016). Nevertheless, carefully designed and interpreted studies would give valuable insight in the underlying neuroplastic mechanisms in MS following rehabilitation.

3 Efficacy of inpatient multidisciplinary rehabilitation in multiple sclerosis: behavioral and fMRI results

Journal of Neurology
https://doi.org/10.1007/s00415-020-09768-6

ORIGINAL COMMUNICATION

Efficacy of inpatient personalized multidisciplinary rehabilitation in multiple sclerosis: behavioural and functional imaging results

Priska Zuber[1,3] · Charidimos Tsagkas[2,3] · Athina Papadopoulou[2,4] · Laura Gaetano[9] · Manuel Huerbin[3] · Emanuel Geiter[2] · Anna Altermatt[3,5] · Katrin Parmar[2] · Thierry Ettlin[6] · Corina Schuster-Amft[6] · Zorica Suica[6] · Hala Alrasheed[7] · Jens Wuerfel[3,4,5] · Jürg Kesselring[7] · Ludwig Kappos[2] · Till Sprenger[2,8] · Stefano Magon[2,3]

Received: 20 June 2019 / Revised: 23 January 2020 / Accepted: 19 February 2020

Abstract

Background Although multidisciplinary rehabilitation programs are commonly used in clinical practice for patients with multiple sclerosis (MS), they are currently underexamined.

Objective This study aims to investigate the efficacy and underlying brain mechanisms of an inpatient multidisciplinary rehabilitation.

Methods Twenty-four patients with relapse-onset MS underwent a 4-week personalized inpatient multidisciplinary rehabilitation and three assessment sessions including MRI, clinical, cognitive and motor function evaluation. Twenty-four healthy controls underwent two assessment sessions 4 weeks apart. Test performances were compared using repeated measures ANOVA, Tukey and *t* tests. A motor sequence learning (MSL) task was presented during fMRI and data were analysed using FSL.

Results Patients had less perceived fatigue, improved walking speed and quality of life following the rehabilitation, which could be maintained at follow-up 4 weeks after rehabilitation. After rehabilitation, differences in accuracy of the MSL task between groups diminished, indicating an improved performance in patients. Improved accuracy went along with changes of brain activity in the left cerebellum and right frontal lobe post-rehabilitation, which could be maintained at follow-up. No changes between sessions were observed in controls.

Conclusion Multidisciplinary rehabilitation may improve highly impacting symptoms through more efficient recruitment of brain regions and therefore positively influence MS patients' quality of life.

Keywords Multidisciplinary rehabilitation · Multiple sclerosis · fMRI · Fatigue · Motor skills · Quality of life

Introduction

MS is the most common inflammatory and neurodegenerative disorder of the central nervous system in early adulthood causing a variety of motor, cognitive and psychological

Electronic supplementary material The online version of this article (https://doi.org/10.1007/s00415-020-09768-6) contains supplementary material, which is available to authorized users.

✉ Stefano Magon
stefano.magon@usb.ch

1 Division of Cognitive Neuroscience, Faculty of Psychology, University of Basel, Basel, Switzerland

2 Department of Neurology, University Hospital Basel, Neurologic Clinic and Policlinic, Petersgraben 4, 4031 Basel, Switzerland

3 Medical Image Analysis Center (MIAC AG), Basel, Switzerland

4 Neurocure Clinical Research Center, Charité–Universitätsmedizin Berlin, Corporate Member of Freie Universität Berlin, Humboldt-Universität Zu Berlin, and Berlin Institute of Health, Berlin, Germany

5 Department of Biomedical Engineering, University of Basel, Basel, Switzerland

6 Reha Rheinfelden, Rheinfelden, Switzerland

7 Department of Neurology and Neurorehabilitation, Clinics of Valens, Valens, Switzerland

8 Department of Neurology, DKD Helios Klinik Wiesbaden, Wiesbaden, Germany

9 F. Hoffman-La Roche Ltd, Basel, Switzerland

Published online: 02 March 2020

symptoms [1, 2]. This polysymptomatic clinical picture is common in MS and associated with reduced quality of life (QoL) [3]. Although disease-modifying therapies showed to reduce the rate of disease progression the progression the development of effective rehabilitation programs are still crucial in the management of the disease.

In this regard, previous studies showed efficacy of symptom-specific rehabilitation programs. It has been reported that out- and inpatient motor rehabilitation can be effective not only in improving mobility, muscle strength and aerobic capacity but also in demonstrating positive effects in treating fatigue [4–6]. Besides, cognitive trainings have been shown to reduce cognitive impairments in MS [7]. Moreover, by using functional magnetic resonance imaging (fMRI), it has been shown that symptom improvement through rehabilitation goes along with changes of brain activity [8, 9]. Indeed, studies on cognitive rehabilitation showed increases in post-training brain activation in the cerebellum, frontal and parietal lobe in patients with MS [10]. Regarding motor deficits only few studies investigated the brain mechanisms of specific motor training and showed controversial results: during a dynamic movement task after outpatient physiotherapy [11] or following a motor training of the upper extremities [12] no task related changes in brain activation were found versus an increased lateralization showing normalized activation in the left hemisphere and cerebellum was described [13, 14]. In contrast, after an inpatient training on global physical functioning, resistance and endurance, a decrease of brain activation in response to a lower limb movement task during the scan was described in a fMRI study [15].

However, in clinical practice, rehabilitative programs for patients with MS do not only focus on single-symptom trainings, but rather combine multiple treatment modalities in order to address the complex symptomatology [2]. The personalized multidisciplinary rehabilitation (PMDR) approach is characterized by a biopsychosocial model and has been defined as "[…] the co-ordinated delivery of intervention by two or more disciplines […]" [2]. In two reviews addressing different settings for rehabilitation in people with MS, "moderate-quality evidence" for long-term improvements in participation, QoL and reducing disability following inpatient PMDR have been reported [6, 16]. Furthermore, improvements in motor performance [2] and reduced fatigue [17] have been described following PMDR in patients with MS.

Although the symptomatology of MS should be seen in a polysymptomatic context, only ten out of 528 studies summarized in the review by Khan and Amatya [6] investigated inpatient PMDR. Based on their findings, the authors concluded that further research is likely to have an impact in the investigation of inpatient PMDR [6]. In addition, previous studies on neuroplastic mechanisms involved in rehabilitation in MS focused on symptom specific rehabilitative strategies. Hence, there is a lack of studies that investigate the efficacy of inpatient PMDR among clinical measures in combination with the underlying brain activity changes in patients with MS.

Therefore, this observational study aimed to address this knowledge gap by (1) investigating the efficacy of an inpatient PMDR as defined in clinical practice on behavioural outcomes including motor and cognitive measures and (2) studying brain mechanisms underpinning potential improvements by detecting task-related BOLD changes in patients with MS compared to an age- and sex-matched population of healthy adults. We hypothesized that PMDR can induce improvement in the performances and as well as changes in the brain activity.

Methods

Participants

Twenty-four patients with MS (16 females; mean age: 47.7 ± 10.1 years; median/range Expanded Disability Status Scale (EDSS [18]): 5 (1.5–6.5); mean disease duration: 15.2 ± 8.8 years) and 24 healthy sex- and age-matched subjects (16 females; mean age: 45.0 ± 10.1 years) were included. Inclusion criteria for patients were: (1) Relapse onset MS [19] (2) age: 18–65 years; (3) EDSS below seven; (4) right-handed; (5) relapse free period > 3 months; (6) unchanged therapy for at least 2 months at the time of inclusion. Patients were allowed to continue their regular immuno-modulatory and symptomatic medical treatments. Controls were included if they were (1) right-handed; (2) no regular medication intake. Exclusion criteria were for both groups any contraindication for MRI and no current or prior brain trauma, psychiatric or neurological disorders (other than MS for the patient group). Written informed consent was given by all participants after detailed explanation of the study procedures. The study was approved by the local ethics committee (Ethikkommission Nordwest- und Zentralschweiz) and was conducted in concordance with the Declaration of Helsinki.

Study design

A longitudinal parallel group design was employed. Eligibility criteria were verified and written informed consent was obtained in each participant. After recruitment, the MS group underwent three assessment sessions including clinical, cognitive, motor evaluation and MRI. All patients underwent an inpatient PMDR program, by spending in average 3.1 weeks (SD 0.51 weeks) in the rehabilitation centre. The first assessment session was performed in the week before entering the rehabilitation, the second within the week following the rehabilitation (in average

32.54 ± 10.84 days between pre and post-rehabilitation sessions), and the third 4 weeks after the rehabilitation. Healthy controls performed two assessment sessions 4 weeks apart (in average 25.77 ± 7.41 days between both sessions), including MRI and cognitive assessment.

Intervention

Rehabilitation programs

All patients underwent an inpatient PMDR program with a mean duration of 16.6 ± 3.2 days (min = 10, max = 25.5 days, excluding weekends) and an average of 46.1 ± 15.3 h (min = 26.5, max = 80.6 h) of rehabilitative activities during this period. The rehabilitation took place in either the Clinics of Valens or the Reha Rheinfelden, both in Switzerland. Following an interdisciplinary approach, the personalized rehabilitation programs were based on the International Classification of Functioning, Disability and Health [20] considering the most relevant factor for MS patients [21]. Patient-specific objectives were identified together with a physiotherapist and an occupational therapist at the day of admission and the goals of the rehabilitation were adapted according to the patients' progress during the rehabilitation. Training settings varied and included both individual and group therapy with a main focus on motor rehabilitation (Table 1).

Measurements

Clinical data

Medical history was taken and an EDSS assessment (18; neurostatus, https://www.neurostatus.net) was performed by trained neurologists at the Department of Neurology, University Hospital Basel.

Neuropsychological assessment

Cognitive functions, specifically attention, procession speed and working memory were evaluated at each assessment session using both paper/pencil and computerized tests. The following tests were administered: Paced Auditory Serial Addition Task (PASAT [22]), oral version of the Symbol Digit Modalities Test (SDMT [23]), forward and backward digit span (WAIS-IV [24]) and Corsi Block Tapping Test [25]. Both groups completed a health-related QoL questionnaire (SF-12 [26]). A scale for fatigue (fatigue scale for motor and cognitive function; FSMC [27]) was administered to the MS patients. The assessments were performed at the Department of Neurology, University Hospital Basel by trained psychologists.

Motor assessment

In both groups, manual dexterity and upper extremity functions were assessed using the 9-Hole Peg Test (9-HPT [28]). Patients underwent a timed 25 foot-walk test (T25-FW [29]) in order to assess mobility and lower extremity function. Two patients did not perform the T25-FW pre-rehabilitation and were therefore excluded in the analyses.

Image acquisition

Functional and structural MRI data were collected at the University Hospital Basel on a 3 T Magnetom Prisma MRI scanner (Siemens Healthineers, Erlangen, Germany) using a dedicated 64-channel dedicated head-coil. The MRI

Table 1 Description and average hours of performed patient-specific rehabilitative therapies

Therapy	Physiotherapy	Occupational therapy	Neuropsychological therapy	Cognitive rehabilitation
Description	Movement therapy Aquatherapy Manual therapy Balance training Incontinence therapy Lokomat therapy Hippotherapy Medical training therapy Jacobson's relaxation Yoga/Pilates Terrain training Ergometer training	Robot-assisted arm training (Armeo) Writing training Garden group Workshop group Relaxation group Recreational therapy Cooking Group	Energy Management and self-management	Cognitive self-training and supervised training of memory, attention and executive functions
Hours of training (M ± SD)	32.8 (± 8.6)	4.5 (± 4.6)	5.3 (± 4.3)	1(± 1.7)

This is a selection of the most frequent therapies. As the rehabilitative programs were selected individually, this selection is not extensive. An extensive list of all therapies and explanation to the specific interventions are reported in the supplementary material III protocol included 3D T1-weighted (T1w) MPRAGE (isotropic resolution of 0.7 mm, TR = 2400 ms, TE = 2.32 ms, TI = 1100 ms, 256 sagittal slices) and high resolution T2-weighted (T2w) imaging (isotropic resolution of 0.7 mm, TR = 3200 ms, TE = 566 ms, 256 sagittal slices). Moreover, multi-band accelerated Echo Planar Imaging (EPI) was used for task-related fMRI with high spatial and temporal resolution (T2*-weighted, isotropic resolution of 2 mm, 72 slices aligned to the AC-PC line; TR = 768 ms; TE = 37 ms; multi-band accelerator factor = 8; measurements = 657).

fMRI task

Both groups performed a motor sequence learning task (MSL; [30]) during each MRI session. It has been shown that the ability of Motor Sequence Learning (MSL) represents a hallmark of normal motor function [31] and that people with MS show impairments in acquiring sequential motor skills [32]. Furthermore, it has been described that cognitive processes are involved in MSL [33] and, therefore, using a MSL task could display a suitable paradigm for detecting changes in brain activation after PMDR. The MSL task included a repeated condition, in which the sequence of movements was identical for each repetition of the condition, and a random condition, in which the sequences of movements changed in each presentation (detailed description in Supplementary Material II). Before the fMRI task, each subject performed a short training session inside the scanner. Reaction time (RT) and accuracy were recorded using a 5-button fibre optic response system (Celeritas; Psychological Software Tools, Pittsburgh, PA). E-Prime (Psychological Software Tools, Pittsburgh, PA) was used to present the stimuli, to synchronize the MRI acquisition with the task and to record the subjects' performances.

Statistical analyses

Behavioural data analyses

Neuropsychological data and task fMRI (reaction time and accuracy) were analysed using two factor mixed design analyses of variance (ANOVA). Significant effects in the mixed design ANOVA were investigated using post hoc Tukey tests. Within group analyses were conducted using paired *t* tests. All data were analysed in R Studio [34].

fMRI data analyses

The fMRI data were pre-processed and analysed using the high-quality model-based fMRI analysis tool FEAT implemented in the FMRIB Software Library v5.0 (FSL [35]). Detailed description of the MRI data processing is reported in the supplementary materials.

In the within-subject analysis, a boxcar function was used to model each block of the two conditions (repeated and random), and then convolved with a double-gamma hemodynamic response function in order to create two explanatory variables (EV), corresponding to the two conditions including 15 blocks each. For the comparisons between the two conditions (repeated versus random and random versus repeated), the estimated head motion parameters were included as nuisance variables in the statistical model. Then, the single subject maps obtained in the within-subject analysis were used as input for a two-sample paired *t* test performed to compare the brain activity between different sessions at group level.

All group analyses were performed using FLAME 1 (Local Analysis of Mixed Effects [36]) and results were corrected for multiple comparisons using cluster-based correction (cluster forming threshold $Z > 2.3$, cluster extent significance threshold of $p < 0.05$).

Results

Behavioural results

In patients we found a significant difference in the T25FW test ($t(19) = 3.12$, $p < 0.05$, Fig. 1) and the motor component of the FSMC ($t(19) = 2.34$, $p < 0.05$, Fig. 2) between pre- and post-rehabilitation. No significant changes were found

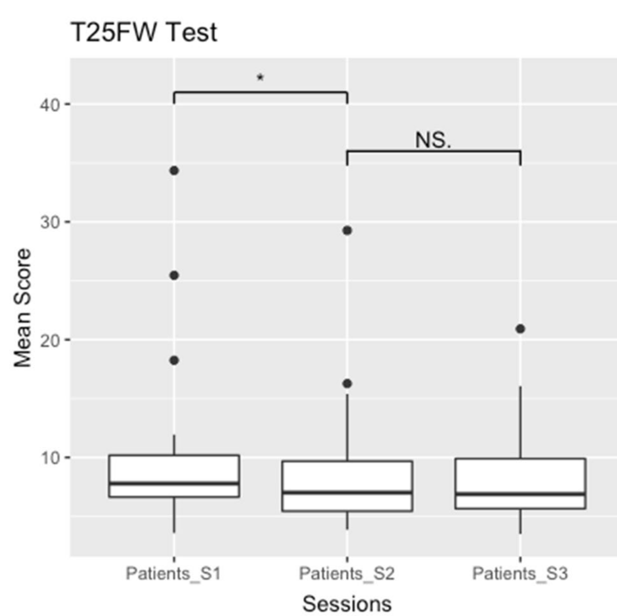

Fig. 1 Significant difference in the T25FW test in the patient group pre- and post-rehabilitation ($p < 0.05$). No significant difference was detected when comparing post-rehabilitation and follow up

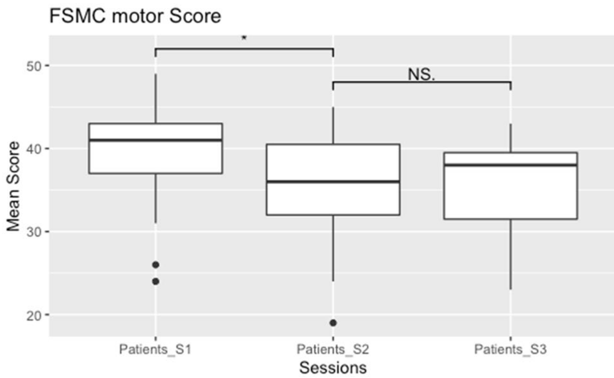

Fig. 2 Significant difference in the FSMC motor score in the patient group pre- and post-rehabilitation ($p<0.05$). No significant difference was detected when comparing post-rehabilitation and follow up

between post-rehabilitation and follow-up in patients and for the cognitive and total score of the FSMC.

For the physical score of the SF-12 questionnaire we found a main effect for groups (patients and controls; $F(1, 37)=126.28$, $p<0.001$), sessions (pre- and post-rehabilitation; $F(1, 37)=7.72$, $p=0.008$) and the interaction between groups and sessions ($F(1, 37)=5.27$, $p=0.02$). Post hoc tests showed that patients had a significant higher SF-12 physical score in post- than pre-rehabilitation, ($t(16)=-2.60$, $p=0.019$). Patients had a lower physical score in sessions 1 ($t(39)=10.54$, $p<0.001$) and 2 ($t(41)=10.59$, $p<0.001$) compared with healthy subjects (Fig. 3). No significant differences between groups or between sessions were found for the mental score of the SF-12. We found no significant differences between post-rehabilitation and follow-up in patients for both, mental and physical score of the SF-12.

In terms of the 9HPT of the right hand a significant effect for groups ($F(1,40)=12.64$, MSE $=90.27$, $p<0.05$) and sessions ($F(1,40)=4.60$, MSE $=10.88$, $p<0.05$) was found. Post hoc tests showed a significant difference between the groups ($p<0.05$) but no significant difference between sessions. For the 9HPT left hand, we found a significant effect for groups ($F(1,40)=1.08$, MSE $=131.49$, $p<0.05$). The post hoc test showed a significant difference between groups ($p<0.05$) for the 9HPT. Further, a significant effect for session in the PASAT ($F(1,40)=4.9$, MSE $=25.26$, $p<0.05$) and in the digit span ($F(1,40)=6.7$, MSE $=2.43$, $p<0.05$) was found. No significant differences between sessions in both PASAT and digit span were found post hoc. No significant effects were found for the Corsi block test and the SDMT. All means and standard deviations are displayed in Table 2.

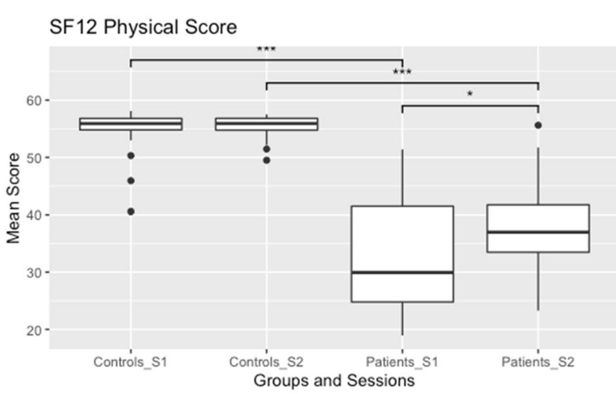

Fig. 3 Significant differences in the SF12 physical score in the patient group pre- and post-rehabilitation ($p<0.05$) and comparing the patient group with the healthy control group at both sessions ($p<0.001$)

Table 2 Means (M) and standard deviation (SD) for performance at session 1 (S1) and session 2 (S2) in the patients and controls and session 3 (S3) in patients

	S1 (M ± SD)	S2 (M ± SD)	S3 (M ± SD)
Patients			
9HPT right hand (s)	27.05 ± 10.48	25.13 ± 8.75	24.12 ± 8.07
9HPT left hand (s)	27.49 ± 10.82	26.86 ± 11.80	25.62 ± 13.16
PASAT	44.1 ± 10.83	45.94 ± 12.31	48.57 ± 9.72
SDMT	49.57 ± 17.47	49.43 ± 17.76	46.91 ± 22.20
Corsi block	14.14 ± 3.34	13.86 ± 3.79	13.48 ± 2.73
Digit span	14.57 ± 3.56	15.67 ± 3.28	15.13 ± 3.17
SF-12 Physical Score	31.39 ± 9.31	36.89 ± 7.85	34.81 ± 9.93
SF-12 Mental Score	51.16 ± 9.10	52.57 ± 8.02	53.79 ± 9.58
FSMC Total Score	71.38 ± 14.77	64.95 ± 15.15	63.30 ± 14.01
FSMC Motor Score	38.76 ± 6.79	34.82 ± 6.57	34.43 ± 5.74
FSMC Cognitive Score	32.62 ± 9.12	30.14 ± 9.95	28.87 ± 9.49
T25FW (s)	10.10 ± 7.44	9.17 ± 5.88	8.95 ± 4.8
Controls			
9HPT right hand (s)	19.30 ± 3.02	18.14 ± 2.59	–
9HPT left hand (s)	19.26 ± 2.91	18.64 ± 2.12	–
PASAT	47.38 ± 8.49	50.38 ± 6.63	–
SDMT	54.81 ± 8.23	59.43 ± 12.04	–
Corsi block	13.62 ± 3.29	14.57 ± 3.83	–
Digit span	14.76 ± 3.11	15.43 ± 3.46	–
SF-12 Physical Score	54.53 ± 4.10	55.31 ± 2.19	–
SF-12 Mental Score	54.98 ± 3.95	55.09 ± 3.67	–
FSMC	–	–	–
T25FW (s)	–	–	–

9HPT nine hole peg test, *PASAT* paced auditory serial addition task, *SDMT* symbol digit modalities test, *SF-12* health related quality of life questionnaire, *FSMC* fatigue scale for motor and cognitive function, *T25FW* timed 25 foot walk test

Task-fMRI results

Accuracy

For both the repeated ($F(1, 38) = 8.02$, MSE = 18.18, $p < 0.05$) and random condition ($F(1, 38) = 5.42$, MSE = 22.60, $p < 0.05$), a significant effect for group was found (Fig. 4). The post hoc test showed a significant difference between the patients and controls ($p < 0.05$). *T* tests between groups at both sessions showed a significantly higher number of mistakes in the repeated ($t(27) = 2.8$, $p < 0.001$) and random condition ($t(33) = 2.8$, $p < 0.001$) of the MSL task at baseline in patients compared to controls at baseline. Interestingly, no significant differences between the groups in the number of mistakes were found post rehabilitation. Comparing post-rehabilitation and follow-up, no significant differences were found in accuracy in the training or the random condition in patients.

Reaction time

In order to consider the learning effect over the conditions during the task, the difference between the first and the last block was built (delta). The difference was then taken as the dependent variable. For the repeated condition, a significant effect for session ($F(1, 38) = 5.24$, MSE = 6531.88, $p < 0.05$) was found. A post hoc analysis of this effect showed no differences between sessions. For the random condition, no significant effects were found.

Brain activity (fMRI data)

In patients, a significant decrease in brain activation was found in the left cerebellum and the right prefrontal lobe when comparing the training and random condition of the MSL task pre- and post-rehabilitation (Table 3, Fig. 5). No difference in brain activation was found between post-rehabilitation and follow-up at 4 weeks in patients. No difference between sessions in the control group and conditions was observed.

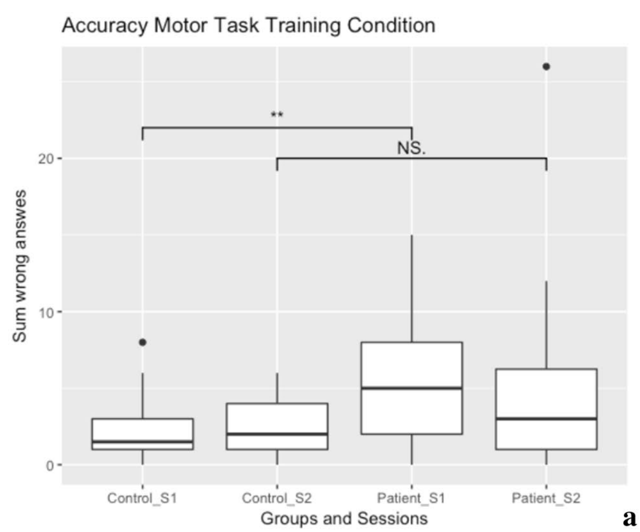

Fig. 4 Means of wrong answers in the MSL task training (**a**) and random (**b** condition in patients and controls at session 1 and session 2 (for patients S1 = pre- and S2 = post rehabilitation). At baseline, the groups differed significantly in both conditions. At session 2 (after rehabilitation), no differences were found. Significant differences are marked with an asterisk

Table 3 Activated brain regions for the difference between the training and random condition of the MSL task comparing pre-and post-rehabilitation in the patient group

Subjects	Brain regions	Number of voxels	*Z*-Max: MNI coordinates *x, y, z* (mm)	*Z*-Max
MS Patients	Left cerebellum	150	42, 30, 44	3.17
	Right prefrontal lobe	221	− 42, − 86, − 4	3.17

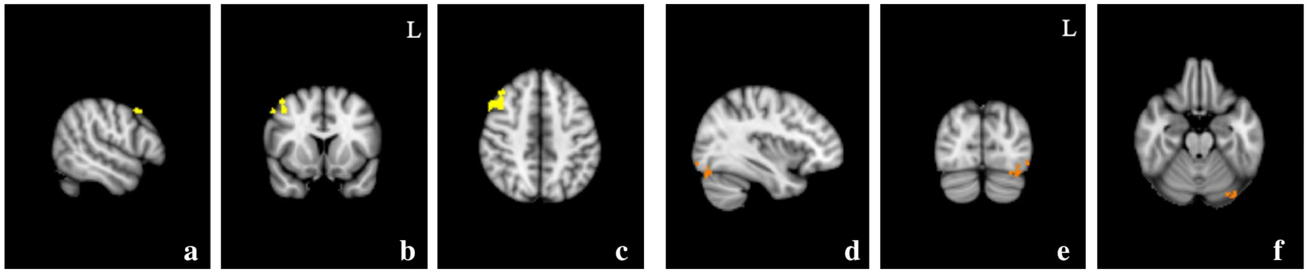

Fig. 5 Significant brain activation (in colour) in the right prefrontal lobe (**a**–**c**) and the left cerebellum (**d**–**f**) in the MS patients when comparing the difference of the training and random condition of the MSL task pre- and post-rehabilitation. Data were corrected for multiple comparisons ($p < 0.05$) and T values ranged between 2.3 and 3.1

Discussion

In this study we investigated the efficacy of an inpatient PMDR program on behavioural outcomes and the underlying brain mechanisms in patients with MS compared to age- and sex-matched healthy adults. Our results show a reduction in perceived fatigue, an improvement of walking abilities and an improvement of health-related QoL after the inpatient PMDR in patients. The improvements were persistent at a 4-week follow-up. We further observed an improved performance on an MSL task in terms of accuracy in patients post rehabilitation, which was maintained at follow-up. Improved accuracy on the MSL task went along with a reduced difference of brain activity between the two conditions (repeated and random) in the left cerebellum and right prefrontal lobe post-rehabilitation in patients, which was also maintained 4 weeks after the rehabilitation. No changes between sessions were observed in healthy controls.

Fatigue and impaired walking ability can be considered as the most common and impacting symptoms of MS [37]. Through limiting coping abilities and participation, fatigue can negatively affect domains of daily living such as employment, socialization or adaption to the disease [38]. Impaired walking ability has not only been reported to reduce the patients, but also the caretakers QoL, wherefore the need of treating walking difficulties in patients has been emphasized [39]. Our study showed improvements on both fatigue and walking ability after a 4-week inpatient PMDR in patients with MS. On the one hand, this result underlines the importance of rehabilitative efforts in treating both symptoms and on the other hand a possible relationship between the symptoms could be assumed. Indeed, a relationship between fatigue and walking ability has been elaborated in a recent study [40]. Furthermore, both symptoms have been described as key determinants of QoL in patients with MS [41, 42] and previous findings showed a positive effect of physical exercise on fatigue [43]. Accordingly, patients in our study reported an improvement in the physical health-related QoL after the PMDR, which has as well been shown in previous investigations applying exercise training [4] and PMDR [44]. Next to this empirical evidence suggesting an association between walking ability and fatigue, it has been theoretically described that walking difficulties result from a combination of common symptoms in MS including fatigue [39]. Furthermore, fatigue has likewise been considered as a complex and multi-factorial symptom and the essential role of a PMDR approach in the management of fatigue has been underlined [39, 45]. However, the applicability of an inpatient PMDR in reducing fatigue in patients with MS has been rarely investigated and our results, therefore, extend these previous findings.

To our best knowledge, the underlying brain mechanisms of inpatient PMDR in patients with MS have not been studied before. We targeted this gap of knowledge by investigating the brain activation changes during an MSL task before and after the PMDR using fMRI. On the behavioural level, we observed an improved performance on the MSL task in patients after the rehabilitation. Although the MSL task was not specifically trained, the initial difference in accuracy between groups diminished after the rehabilitation, indicating an improvement in performance for the patients. On the neurofunctional level, our fMRI data indicate that the higher accuracy on the MSL task went along with a reduced difference between conditions in the left cerebellum and right prefrontal cortex after the PMDR in patients, whereas no brain activation changes between the two sessions were observed in controls. The involvement of the cerebellum in MSL and more specifically its role in accuracy through a relationship between decreases in errors and left cerebellar activity has been described previously [46, 47]. Additionally, it has been described that only cerebellar areas were the main common brain regions between activation patterns of temporal accuracy in a motor task performance and self-perceived fatigue [48]. Indeed, without applying rehabilitation an increased activation in cerebellar and prefrontal regions was found in patients with high self-perceived fatigue. The decrease in errors in the MSL task, self-perceived fatigue and brain activity in the left cerebellum and prefrontal regions following the inpatient PMDR found in the present study strengthens the involvement of prefrontal and cerebellar regions in MSL and fatigue and could further indicate a recovery pattern induced by the PMDR. Nevertheless, studies targeting brain activation changes after specific motor training using task-fMRI showed controversial results and indicated that target-selected trainings rather than holistic approaches lead to enhanced neuroplasticity [9]. A study on healthy subjects revealed a redistribution of functional brain activation after the motor task training, whereas increased and as well bilaterally decreased responses have been reported [47, 49]. In MS patients, no brain activation changes have been described following a 30-min upper extremity motor training [12] or a 5-week inpatient motor rehabilitation [11]. Only one recently published study that investigated the effect of motor rehabilitation in lower limbs using task-related fMRI on foot movements of plantar dorsiflexion and the underlying brain activation showed a reduced peak activity in the contralateral primary motor cortex, which was interpreted as a recovery pattern by the authors [15]. The decrease of task-related brain activity after the PMDR found in our study further supports a recovery pattern and may, therefore, indeed reflect a more efficient recruitment of brain regions and by that mirror the efficacy of an inpatient PMDR in improving MS-related symptoms.

Finally, we found no difference in walking ability, fatigue, QoL and MSL task performance and brain activation between post-rehabilitation and follow-up, indicating that the changes induced by the rehabilitation were maintained at a short-term follow-up. Even though it is of utmost importance that rehabilitative efforts can be maintained over time, previous literature showed varying evidence for long-term effects which are dependent on rehabilitation settings and the effect on symptoms [44]. Therefore, our results support the efficacy of clinically defined PMDR and their importance for patients in reducing the disease-related burden as a result of the polysymptomatic presentation of MS. However, future studies should additionally consider not only short- but also long-term follow-up periods to assess the efficacy of PMDRs.

In our study, no differences between groups could be found in the neuropsychological testing. One reason for this finding could be that the patients in our sample experienced a higher amount of physical impairment, mobility restrictions or fatigue rather than cognitive impairment. This may be displayed in the self-reported quality of life, where the patients significantly differed in their scores on the physical subscale to the healthy controls at both sessions, but showed the same levels of mental health-related quality of life compared to healthy controls at both sessions. This may indicate that the patients felt less restricted by the cognitive symptoms in their daily life. Furthermore, no changes between sessions in the neuropsychological testing could be observed. Although the MD rehabilitation in our study did include in average 1 h of cognitive rehabilitation in total, the focus of the program was on physical and occupational trainings (Table 1) and, therefore, less change in cognitive measures can be expected.

The small sample size and the lack of a placebo group could be considered as a limitation. However, it is nearly impossible to create an appropriate active control condition for a PMDR program. Therefore, we included an age- and sex-matched sample of healthy adults to control for practice effects in both behavioural and MRI assessments. An additional limitation is that the current investigated inpatient PMDR program focused primarily on motor rehabilitation. Thus, the described results and improvements may not generalize to distinct functions such as cognition. Finally, future studies should investigate which group of patients may be more likely to benefit from PMDR as presented here based on their type of impairment level or predominant symptom.

In conclusion, this study suggests that the PMDR approach should be considered in the treatment of fatigue and walking abilities since it has the possibility to target and improve two highly impacting symptoms at the same time and by that positively influence participation in daily activities and so patients' QoL. Among one of the first, this study additionally provides evidence on the underlying mechanisms of PMDR by suggesting a more effective recruitment of brain regions in patients with MS. Therefore, this study provides evidence that rehabilitative efforts of a PMDR approach may be detected on both the clinical and neurofunctional level.

Acknowledgement This work was supported by the Swiss Multiple Sclerosis Society (SMSS) [Grant Number 2015]. We want to thank all participants and the SMSS for supporting this project.

Compliance with ethical standards

Conflicts of interest Zuber, P., Tsagkas, C., Huerbin M., Geiter, E., Altermatt, A., Suica, Z., Alrasheed H. and Kesselring, J. have nothing to disclose. Papadopoulou A.: consulted for Teva, received speaker-fee from Sanofi-Genzyme, travel support from Bayer AG, Teva, UCB-Pharma AG, Roche and research support from the University of Basel and the Swiss National. Science Foundation. Gaetano, L.: former employee of Novartis is currently working for F. Hoffmann-La Roche Ltd. Parmar, K.: holds a grant of the Baasch -Medicus foundation. Her institution (University Hospital Basel) received speakers honoraria from Novartis and ExceMED and travel support by Novartis Switzerland. Ettlin, Th.: Institution (Reha Rheinfelden) received and used exclusively for research support: consultancy fee and speaker fees from Grünenthal; support of educational activities Almirall, A. Menarini, Allergan, Bayer, Biogen, Bristol-Myers Squibb SA, Daiichi-Sankyo, Grünenthal, IBSA Pharma AG, Lundbeck, Mepha, Merck, Merz Pharma, MSD Merck Sharp & Dohme AG, Mundipharma, Pfizer, Sanofi Genzyme, Sigma-Tau, Zambon. Schuster-Amft, C.: Institution (Reha Rheinfelden) received and used exclusively for research support from Innosuisse; Swiss National Physiotherapy Association "physioswiss"; European Union; Foundation to promote Science, Research, Educa-

tion, and Health; Frieda Locher Hofmann Foundation; Forschungfonds Canton. Aargau. Wuerfel, J.: CEO of MIAC AG Basel, Switzerland. He served on scientific advisory boards of Actelion, Biogen, Genzyme-Sanofi, Novartis, and Roche. He is or was supported by grants of the EU (Horizon2020), German Federal Ministeries of Education and Research (BMBF) and of Economic Affairs and Energy (BMWI). Kappos, L: Institution (University Hospital Basel) received and used exclusively for research support: steering committee, advisory board, and consultancy fees from Actelion, Addex, Bayer HealthCare, Biogen, Biotica, Celgene/Receptos, Genzyme, Lilly, Merck, Mitsubishi, Novartis, Ono Pharma, Pfizer, Sanofi, Santhera, Siemens, Teva, UCB, Xenoport; speaker fees from Bayer HealthCare, Biogen, Merck, Novartis, Sanofi, Teva; support of educational activities from Bayer HealthCare, Biogen, CSL Behring, Genzyme, Merck, Novartis, Sanofi, Teva; grants from Bayer HealthCare, Biogen, F. Hoffmann-La Roche Ltd, Merck, Novartis, the European Union, the Roche Research Foundations, the Swiss Multiple Sclerosis Society, the Swiss National Research Foundation. Sprenger, T.: Author's current and previous institutions have received compensation for author serving on scientific advisory boards/consultation and speaking from Actelion, ATI, Biogen, Desitin, Electrocore, Novartis, Sanofi, Genzyme, and Teva; research support from Novartis, Swiss MS Society, and Swiss National Science Foundation. Magon, S.: is currently working for F. Hoffmann-La Roche Ltd. He has received research support from Swiss MS Society, Swiss National Science Foundation, University of Basel and Stiftung zur Förderung der gastroenterologischen und allgemeinen klinischen Forschung sowie der medizinischen Bildauswertung University Hospital Basel. He also received travel support from Biogen and Genzyme.

Ethical standards The study was approved by the local ethics committee (Ethikkommission Nordwest- und Zentralschweiz) and was conducted in concordance with the Declaration of Helsinki.

References

1. Thompson A (2001) Symptomatic management and rehabilitation in multiple sclerosis. J Neurol Neurosurg Psychiatry 71:22–27
2. Beer S, Khan F, Kesselring J (2012) Rehabilitation interventions in multiple sclerosis: an overview. J Neurol 259(9):1994–2008
3. Kister I, Bacon TE, Chamot E, Salter AR, Cutter GR, Kalina JT et al (2013) Natural history of multiple sclerosis symptoms. Int J MS Care 15(3):146–156
4. Latimer-Cheung AE, Pilutti LA, Hicks AL, Ginis KAM, Fenuta AM, MacKibbon KA, Motl RW (2013) Effects of exercise training on fitness, mobility, fatigue, and health-related quality of life among adults with multiple sclerosis: a systematic review to inform guideline development. Arch Phys Med Rehabil 94(9):1800–1828
5. Asano M, Berg E, Johnson K, Turpin M, Finlayson ML (2015) A scoping review of rehabilitation interventions that reduce fatigue among adults with multiple sclerosis. Disabil Rehabil 37(9):729–738
6. Khan F, Amatya B (2017) Rehabilitation in multiple sclerosis: a systematic review of systematic reviews. Arch Phys Med Rehabil 98(2):353–367
7. Flavia M, Stampatori C, Zanotti D, Parrinello G, Capra R (2010) Efficacy and specificity of intensive cognitive rehabilitation of attention and executive functions in multiple sclerosis. J Neurol Sci 288(1–2):101–105
8. Flachenecker P (2015) Clinical implications of neuroplasticity: the role of rehabilitation in multiple sclerosis. Front Neurol 6:36
9. Prosperini L, Piattella MC, Giannì C (2015) Functional and structural brain plasticity enhanced by motor and cognitive rehabilitation in multiple sclerosis. Neural Plast 2015:481574
10. Chiaravalloti ND, Genova HM, DeLuca J (2015) Cognitive rehabilitation in multiple sclerosis: the role of plasticity. Front Neurol 6:1–10
11. Rasova K, Krasenksy J, Havrdova E, Obenberger J, Seidel Z, Dolezal O et al (2005) Is it possible to actively and purposely make use of plasticity and adaptability in the neurorehabilitation treatment of multiple scelorsis patients? A pilot project. Clin Rehabil 19(2):170–181
12. Morgen K, Kadom N, Sawaki L, Tessitore A, Ohayon J, McFarland H et al (2004) Training-dependent plasticity in patients with multiple sclerosis. Brain 127(11):2506–2517
13. Bonzano L, Tacchino A, Brichetto G, Roccatagliata L, Dessypris A, Feraco P et al (2014) Upper limb motor rehabilitation impacts white matter microstructure in multiple sclerosis. Neuroimage 90:107–116
14. Bonzano L, Pedullà L, Tacchino A, Brichetto G, Battaglia MA, Mancardi GL et al (2019) Upper limb motor training based on task-oriented exercises induces functional brain reorganization in patients with multiple sclerosis. Neuroscience 410:150–159
15. Tavazzi E, Bergsland N, Cattaneo D, Gervasoni E, Laganà MM, Dipasquale O et al (2018) Effects of motor rehabilitation on mobility and brain plasticity in multiple sclerosis: a structural and functional MRI study. J Neurol 265(6):1393–1401
16. Amatya B, Khan F, Galea M (2019) Rehabilitation for people with multiple sclerosis: an overview of cochrane reviews. Vol. 2019, cochrane database of systematic reviews. John Wiley and Sons Ltd., New Jerssey
17. Romberg A, Ruutiainen J, Puukka P, Poikkeus L (2008) Fatigue in multiple sclerosis patients during inpatient rehabilitation. Disabil Rehabil 30(19):1480–2145
18. Kurtzke JF (1983) Rating neurologic impairment in multiple sclerosis: an expanded disability status scale (EDSS). Neurology 33(11):1444–1444
19. Polman CH, Reingold SC, Banwell B, Clanet M, Cohen JA, Filippi M et al (2011) Diagnostic criteria for multiple sclerosis: 2010 revisions to the McDonald criteria. Ann Neurol 69(2):292–302
20. World Health Organization (2001) International classification of functioning, disability and health: ICF. WHO, Geneva
21. Holper L, Coenen M, Weise A, Stucki G, Cieza A, Kesselring J (2010) Characterization of functioning in multiple sclerosis using the ICF. J Neurol 257(1):103–113
22. Gronwall DMA (1977) Paced auditory serial-addition task: a measure of recovery from concussion. Percept Mot Skills 44(2):367–373
23. Smith A (1982) Symbol digit modalities test. Western Psychological Services, Los Angeles
24. Wechsler D (2008) Wechsler adult intelligence scale-fourth edition (WAIS–IV). The Psychological Corporation, San Antonio
25. Corsi P (1972) Memory and the medial temporal region of the brain. McGill University, Montreal
26. Ware JE Jr, Kosinski M, Keller SDA (1996) 12-Item Short-Form Health Survey: construction of scales and preliminary tests of reliability and validity. Med Care 34(3):220–233
27. Penner IK, Opwis K, Kappos L (2007) Relation between functional brain imaging, cognitive impairment and cognitive rehabilitation in patients with multiple sclerosis. J Neurol 254(2):53–57
28. Mathiowetz V, Weber K, Kashman N, Volland G (1985) Adult norms for the nine hole peg test of finger dexterity. Occup Ther J Res 5(1):24–38
29. Kaufman M, Moyer D, Norton J (2000) The significant change for the timed 25-foot walk in the multiple sclerosis functional composite. Mult Scler J 6(4):286–290

30. Magon S, Pfister A, Laura G, Lüthi M, Papadopoulou A, Kappos L et al (2019) Short timescale modulation of cortical and cerebellar activity in the early phase of motor sequence learning: an fMRI study. Brain Imaging Behav. https://doi.org/10.1007/s11682-019-00167-8
31. Tacchino A, Bove M, Roccatagliata L, Luigi Mancardi G, Uccelli A, Bonzano L (2014) Selective impairments of motor sequence learning in multiple sclerosis patients with minimal disability. Brain Res 1585:91–98
32. Deroost N, Smetcoren S, Vandenbossche J, D'Hooghe M, Kerckhofs E (2014) Implicit and explicit learning of sequential motor skill in multiple sclerosis: directions for rehabilitation. J Neurosci Rehabil 1(1):1–13
33. Seidler RD, Bo J, Anguera JA (2012) Neurocognitive contributions to motor skill learning: the role of working memory. J Mot Behav 44(6):445–453
34. R Core Team R (2013) A language and environment for statistical computing. R Found Stat Comput. R Core Team, Vienna
35. Smith SM, Jenkinson M, Woolrich MW, Beckmann CF, Behrens TE, Johansen-Berg H, Niazy RK (2004) Advances in functional and structural MR image analysis and implementation as FSL. Neuroimage 23:208–219
36. Beckmann CF, Jenkinson M, Smith SM (2003) General multilevel linear modeling for group analysis in FMRI. Neuroimage 20(2):1052–1063
37. Flachenecker P, Kümpfel T, Kallmann B, Gottschalk M, Grauer O, Rieckmann P et al (2002) Fatigue in multiple sclerosis: a comparison of different rating scales and correlation to clinical parameters. Mult Scler J 8(6):523–526
38. Bakshi R (2003) Fatigue associated with multiple sclerosis: diagnosis, impact and management. Mult Scler J 9(3):219–227
39. LaRocca NG (2011) Impact of walking impairment in multiple sclerosis. Patient 4(3):189–201
40. Dalgas U, Langeskov-Christensen M, Skjerbæk A, Jensen E, Baert I, Romberg A, Béthoux F (2018) Is the impact of fatigue related to walking capacity and perceived ability in persons with multiple sclerosis? A multicenter study. J Neurol Sci 387:179–186
41. Wood B, van der Mei I, Ponsonby A-L, Pittas F, Quinn S, Dwyer T et al (2013) Prevalence and concurrence of anxiety, depression and fatigue over time in multiple sclerosis. Mult Scler J 19(2):217–224
42. Ysrraelit MC, Fiol MP, Gaitán MI, Correale J (2018) Quality of life assessment in multiple sclerosis: different perception between patients and neurologists. Front Neurol 8:729
43. Pilutti LA, Greenlee TA, Motl RW, Nickrent MS, Petruzzello SJ (2013) Effects of exercise training on fatigue in multiple sclerosis: a meta-analysis. Psychosom Med 75(6):575–580
44. Khan F, Turner-Stokes L, Ng L, Kilpatrick T (2007) Multidisciplinary rehabilitation for adults with multiple sclerosis. Cochrane Database Syst Rev. https://doi.org/10.1002/14651858.CD006036.pub2
45. Induruwa I, Constantinescu CS, Gran B (2012) Fatigue in multiple sclerosis a brief review. J Neurol Sci 323(1–2):9–15
46. Bernard JA, Seidler RD (2013) Cerebellar contributions to visuomotor adaptation and motor sequence learning: an ALE meta-analysis. Front Hum Neurosci 7:27
47. Dayan E, Cohen LG (2011) Neuroplasticity subserving motor skill learning. Neuron 72(3):443–454
48. Pardini M, Bonzano L, Roccatagliata L, Mancardi GL, Bove M (2013) The fatigue-motor performance paradox in multiple sclerosis. Sci Rep 3(1):2001
49. Floyer-Lea A, Matthews PM (2005) Distinguishable brain activation networks for short- and long-term motor skill learning. J Neurophysiol 94(1):512–518

4 Plasticity in the Context of the Healthy Population

As outlined in the previous sections, plasticity of the brain is the basic concept behind restoring different domains of functioning after brain damage. Besides, plasticity is the cornerstone of normal functioning in healthy people in order to be able to learn and adapt to the environment (Cramer et al., 2011; Willis & Schaie, 2009). Compared to the motor system, which is able to quickly adapt itself to activity-dependent input based on its high flexibility (Horton et al., 2017), plasticity of the cognitive system however is less straight forward. Indeed, process-based cognitive functions such as working memory involve many different processes simultaneously, which has been shown, for example, in its interaction with motor functions (Marvel, Morgan, & Kronemer, 2019). Thus, plasticity will first be reviewed in the light of the healthy population by first introducing the concept of cognitive plasticity followed by the current challenges in the field of cognitive plasticity induced by training in the context of working memory. Furthermore, it has been described that motor behavior is highly dependent on cognitive and perceptual processes (Horton et al., 2017). Nevertheless, the interactive or additive effects between the both in the context of plasticity have been far less investigated. The last original articles aimed at studying the interaction of motor and cognitive plasticity induced by training.

4.1 Cognitive Plasticity

Plasticity can also be understood as the capacity of acquiring new cognitive skills, termed as *cognitive plasticity* (Mercado, 2008). Whereas cognitive skills can be described as functions with the ability to be improved through learning and practice (Willis & Schaie, 2009), the concept of cognitive plasticity has been defined as the " […] capacity for changes in flexibility, that is, the capacity for changes in the possible range of cognitive performance."(Lövdén, Bäckman, Lindenberger, Schaefer, & Schmiedek, 2010). Further, neural plasticity can be understood as the basis of cognitive plasticity (Mercado, 2008). Indeed, neuroplastic mechanisms such as neurogenesis, remodeling and formation of new connections, axonal sprouting and pruning or cell migration are responsible for creating networks in the brain following learning and memory (Von Bernhardi et al., 2017). Additionally, the neurogenesis in adulthood has been suggested to relate to learning in the sense of an enrichment of neural connectivity in the brain (Grafman, 2000). Overall, plasticity in the healthy population focuses on the intraindividual potential in cognitive functions, which can also be understood as the intraindividual range of plasticity (Willis & Schaie, 2009).

In order to explain the individual potential in the sense of a person's neural supply and how plasticity has the possibility to adapt it, a theoretical model has been suggested. According to the *supply-demand model*, neural and cognitive plasticity can only occur if there is a discrepancy between requirements to perform cognitive tasks, so-called environmental "demands" and brain capacity, also termed as functional "supply". This mismatch between supply and demand can either be the result of changes in the environmental demands, e.g., adaptation to a new cognitive task, or, changes in the functional supply, e.g., following brain injury or old age. It has been described that the amount of mismatch in individuals can be assessed using fMRI (Lövdén et al., 2010). Furthermore, the supply-demand model suggests, in order to induce cognitive plastic changes in the healthy population, the duration and magnitude of the supply-demand mismatch needs to be maximized and that cognitive trainings or interventions may have the ability to do so (Lövdén et al., 2010).

It has been outlined that human development is characterized by gains and losses in cognitive functioning at different phases throughout life (Lövdén et al., 2010; Willis & Schaie, 2009). Developmental plasticity does take place as maturation of brain regions due to the acquisition of cognitive functions, such as working memory in childhood and adolescence, with a peak performance at around the age of 20-30 years. Although cognitive functions such as vocabulary, arithmetic or general knowledge increase until the age of 50 years, later adulthood is characterized by an outweighing decline of cognitive functions due to the course of healthy aging (Hartshorne & Germine, 2015). While plasticity in old age has been described as maintenance of functions rather than growth (Kühn & Lindenberger, 2015) and losses overshadow gains with increasing age, cognitive interventions focus on functions that have been described to decline already early in the lifespan. Hence, process-based abilities such as working memory, episodic memory, executive functioning or processing speed are typically targeted with focused training (Nguyen, Murphy, & Andrews, 2019; Willis & Schaie, 2009). In the following sections, cognitive plasticity induced by cognitive training will be discussed in the context of working memory.

4.2 Working Memory

Working Memory (WM) refers to a cognitive system which temporarily maintains, stores and manipulates information and acts as an interface between perception, long-term memory and action (Baddeley, 2003, 2012). Among several theoretical frameworks that have been suggested to describe WM, the multicomponent model (Baddeley, Hitch, & Bower, 1974) is the most investigated and broadly accepted one. Originally, the model consisted of three components. First, the *phonological loop* is responsible for passively storing and maintaining phonological information through rehearsal in order to prevent the acoustic information from decay. Second, the *visuospatial sketchpad* displays a short-time storage for spatial and visual information and is further able to manipulate this information with a limited capacity. Two different systems for visual and spatial information prevent a disturbance or interference in the processing of this information. The phonological loop and the visuospatial sketchpad are also referred to as “slave systems” of the central executive. Indeed, the *central executive* was first proposed as a storage but later seen as a processing and coordinating third component that focuses attention, connects to the long-term memory, shifts between tasks or retrieval strategies and is responsible for inhibitory control. Later, the *episodic buffer* was added to the model, which is described as a multi-modal storage system with a limited capacity that can bind visual or phonological information together to create episodes (Baddeley, 2012).

The limited amount of information that can be stored within WM is also referred to as WM capacity and varies between individuals (Morrison & Chein, 2011). Indeed, the individual WM capacity determines the ability to carry out higher-order cognitive functions such as comprehension, learning, reasoning and decision-making and has thus a substantial influence on everyday life activities in healthy people (Diamond, 2013; Morrison & Chein, 2011). Furthermore, WM is one of the process-based cognitive functions displaying an middle age onset age-dependent decline in the course of healthy aging (Willis & Schaie, 2009). WM decline throughout adult life however seems to show a non-linear trajectory, since WM relevant mechanisms such as inhibition of irrelevant information and processing speed diminish with a late onset (Borella, Ghisletta, & De Ribaupierre, 2011; Nyberg, Lövdén, Riklund, Lindenberger, & Bäckman, 2012). While WM is relevant for a large number of everyday functions, age-related decline or WM impairment in the sense of reduced WM capacity can affect daily functioning or work performances and may thus reduce the quality of life (Nyberg et al., 2012). Indeed, neurological, psychiatric or developmental disorders can lead to mild but also early and severe impairments in WM (Budson & Price, 2007; Redick, 2019).

After being assumed to be strictly limited, it has nowadays been established that WM capacity can be expanded through training in the healthy young and old population (Gathercole, Dunning, Holmes, & Norris, 2019; Ophey, Roheger, Folkerts, Skoetz, & Kalbe, 2020). As a consequence, cognitive trainings on core cognitive functions such as WM have been proposed as a possible tool to increase individual WM capacity, improve performance on learning, problem solving or reasoning and to reduce the impact of cognitive decline (Katz, Shah, & Meyer, 2017; Klingberg, 2010).

4.3 Current State and Challenges of Cognitive Training

The first positive attempts of increasing memory and attention in a sample of school children with targeted cognitive training were mentioned in 1890 and gained huge interest amongst psychologists at that time. However, first replications in the beginning of the 20th century led to controverse results, specifically regarding their improvements on untrained cognitive functions following the trainings (Katz et al., 2017). Ever since, the field of cognitive training emerged and led to a large body of literature specifically testing the efficacy of WM trainings. Almost 100 years later, the publication of first studies on computerized WM trainings resulted in high media attention again, with numerous commercial companies developing “brain training” games. This was mainly based on described improvements in the measures of intelligence in healthy young people following training of WM (Jaeggi, Buschkuehl, Jonides, & Perrig, 2008; Katz et al., 2017; Klingberg et al., 2005). Although initial promising results of those first computerized WM training studies increased the interest in developing WM trainings considerably (Soveri, Antfolk, Karlsson, Salo, & Laine, 2017), the question regarding the efficacy of trainings resulted in the same debate as over hundred years ago. For its understanding it is necessary to outline how efficacy is assessed following cognitive training.

Typically, efficacy is investigated as the improvement after the training on an untrained cognitive task and is termed as *transfer effect* (Jaeggi et al., 2008). Transfer effects can arise as *near transfer*, when improvements on untrained tasks similar to the trained tasks are described, e.g., improvements on a WM task that is structurally similar to the trained WM task. However, it is of much higher interest if improvement on a task of a different cognitive function is detected following cognitive training, also termed as *far transfer* (Katz et al., 2017; Teixeira-Santos et al., 2019). Dependent on the investigated population, far transfer following WM training is typically assessed with tasks on fluid intelligence, while in young individuals also performance on verbal or visual learning or school outcomes (Melby-Lervåg, Redick, & Hulme, 2016) or in old adults measures of daily life activities are investigated (Basak, Qin, & O’Connell, 2020). Meta-analytic reviews on transfer effects following WM training agree on the existence of near transfer in young adults (Melby-Lervåg et al., 2016; Soveri et al., 2017). However, the effects were described to be short-term and limited to the trained task paradigm and no convincing evidence was found for far transfer after training (Melby-Lervåg et al., 2016). In old adults, short- and long-term near transfer (Nguyen et al., 2019; Teixeira-Santos et al., 2019) as well as small far transfer effects to daily functioning were described following WM training (Basak et al., 2020). Although improvement of WM after training has been widely described among the healthy population, the task-specific near transfer and limited evidence for far transfer resulted in a heated debate concerning the efficacy of WM trainings. Indeed, the specificity of learning to the applied tasks has not only been described following WM trainings but displays a major challenge in designing efficient strategies to induce cognitive plasticity following training in healthy adults but also following rehabilitation in pathological conditions (Green & Bavelier, 2008).

The reason for diverging results on transfer effects following cognitive trainings are manifold. First, there are numerous paradigms and activities available with the aim of training cognitive functions. The variability in training activities does not allow to draw conclusions of single studies to the efficacy of cognitive training as a whole (Katz et al., 2017). Regarding WM, the diversity of existing training programs has been described to limit the possibility to compare studies and has been suggested as an explaining factor for controverse transfer effects (Pergher et al., 2020). In addition, comparison between studies is limited through methodological considerations regarding the inclusion of appropriate control condition, accounting for practice effects, consistency in applied transfer measures or dosage and setting of training (Morrison & Chein, 2011; Scharfen, Jansen, & Holling, 2018). So far, only a few studies systematically investigated the comparative efficacy of training paradigms (Holmes, Woolgar, Hampshire, & Gathercole, 2019; Minear et al., 2016). However, this approach may help in targeting the challenge to identify if a given training paradigm may be more effective in enhancing cognitive functions or to differentiate which paradigms target which cognitive construct (Morrison & Chein, 2011).

The question of which cognitive construct is actually trained by the applied training can be seen as the second challenge in cognitive training research. Indeed, current research progresses in the direction of not only testing the efficacy of training, but identifying “what” is trained and for “whom” the possible effects can be of relevance (Katz et al., 2017). In this context, specifically the lack of theory-driven WM training approaches limits the understanding of which underlying mechanisms may induce transfer effects (Katz et al., 2017; Redick, 2019). Further, there is the need to clarify which training tasks share mechanisms with the studied transfer measure in order to detect the relevance of the training for the appropriate population (Redick, Wiemers, & Engle, 2019).

Hence, task-related processes have been described to be associated with improved WM following training (Minear et al., 2016). So has WM performance been described to be limited only when distractive irrelevant information was present in healthy old adults (Gazzaley et al., 2008). Indeed, training on preventing irrelevant information of assessing WM – termed as filtering efficiency – has been shown to improve WM performance to the same extent than WM training (Li, He, Wang, Hu, & Guo, 2017; Schmicker, Schwefel, Vellage, & Müller, 2016). Studies however are needed to determine if and how filtering efficiency displays a task-related process of WM training in healthy old adults.

In addition to the investigation of behavioral processes underlying WM training, the examination of neural correlates may further establish the ongoing processes of transfer effects following WM training (Katz et al., 2017). Indeed, WM has been described to involve a large number of brain areas since it is involved in numerous higher order cognitive functions (Eriksson, Vogel, Lansner, Bergström, & Nyberg, 2015). Besides, recent functional imaging evidence indicates an interaction of WM paradigms with the motor system (Marvel et al., 2019). It has been described that sub-regions of motor structures in the brain are simultaneously participating in motor but also cognitive processes and that varying capacity of WM among individuals can be associated to recruitment of the motor networks in order to maintain WM performance (Marvel et al., 2019).

It can thus be summarized, that the former question regarding the efficacy of cognitive training is recently being replaced by the need of studying what mechanisms are trained, under which conditions they might work and what processes may be related to training and outcome tasks in order to induce transfer effects. By targeting those questions, it can be understood how cognitive training has the possibility to adapt the individual cognitive potential and thus induce cognitive plasticity. In two studies, we aimed at overcoming the described challenges by 1) investigating a theory-driven WM training with and without inclusion of distractor inhibition as possible task-related process in comparison to other WM training strategies in a methodologically well-designed clinical trial in healthy old adults (**B**) and 2) studying the additive and interactive neural and behavioral mechanisms induced by motor and WM training by applying functional connectivity measures (**C**).

5 Original Research on Underlying Mechanisms of Working Memory Training

5.1 Development of a Model-based Working Memory Training with and without Distractor Inhibition and Investigation of its Comparative Efficacy

Investigation of a Model-based Working Memory Training with and without Distractor Inhibition and its Comparative Efficacy: A Randomized Controlled Trial on Healthy Old Adults

Priska Zuber[1,2*], Emanuel Geiter[3, 5], Dominique J.-F. de Quervain[1,2, 4], Stefano Magon[5,6*]

[1]Division of Cognitive Neuroscience, Faculty of Psychology, University of Basel, Switzerland

[2]Transfaculty Research Platform, University of Basel, Basel, Switzerland

[3]Medical Image Analysis Center (MIAC AG), Basel, Switzerland

[4]University Psychiatric Clinics, University of Basel, Basel, Switzerland

[5]Department of Neurology, University Hospital Basel, University of Basel, Switzerland

[6]Roche Pharma Research and Early Development, Roche Innovation Center Basel, F. Hoffmann-La Roche Ltd., Basel, Switzerland

*** Correspondence:**
Dr. phil. Stefano Magon (stefano.magon@roche.com) and MSc Priska Zuber (priska.zuber@unibas.ch)

Keywords: Working Memory, Training, Filtering Efficiency, Distractor Inhibition, Cognition, Aging, Cognitive Training, Practice Effects

Abstract

Background: Various working memory (WM) trainings have been tested, but differences in experimental designs, lack of theoretical background and need of identifying task-related processes such as filtering efficiency limit conclusions about their comparative efficacy.
Objectives: In this study we compared the efficacy of a model-based WM training with (MB^+) and without (MB) distractor inhibition on improving WM capacity to a dual-n-back and active control condition.
Methods: This randomized clinical trial included 123 healthy elderly adults (78 women, 45 men; aged 64.1±8.3 years). All groups underwent twelve 40-min training sessions over three weeks and four cognitive testing sessions. The first two sessions served as double-baseline to account for practice effects. Primary outcome was WM capacity post-training measured by complex span tasks. Near and far transfer was assessed by simple span, n-back, visuospatial and verbal learning, processing speed and reasoning tasks.
Results: Due to preliminary termination (COVID-19) 93 subjects completed the post-training and 60 the follow-up session. On whole group level, practice-effects occurred from pre-baseline to baseline in WM capacity ($b = 4.85$, $t(103) = 4.01$, $p < .001$, $r = 0.37$). Linear mixed effects models revealed a difference in WM capacity post-training between MB^+ and MB ($b = -9.62$, $t(82) = -2.52$), $p = .014$, $r = 0.27$) and a trend difference between MB^+ and dual-n-back ($b = -7.59$, $t(82) = -1.87$), $p = .065$, $r = 0.20$) and control training ($b = -7.08$, $t(82) = -1.86$), $p = .067$, $r = 0.20$). Univariate analyses showed an increase between pre- and post-training for WM capacity within MB^+ ($t(22) = -3.34$, $p < 0.05$) only. There was no difference between groups

pre- and post-training regarding near and far transfer. Univariate analyses showed improved visuospatial learning within MB^+ ($t(21) = -3.8$, $p < .05$), improved processing speed ($t(23) = 2.19$, $p < .05$) and n-back performance ($t(23) = 2.12$, $p < .05$) in MB and improved n-back performance ($t(25) = 3.83$, $p < .001$) in the dual-n-back training.
Interpretation: A model-based WM training including filtering efficacy may be a promising approach to increase WM capacity and needs further investigation in randomized controlled studies.

1 Introduction

Working Memory (WM) refers to a cognitive system which temporarily maintains, stores and manipulates information and acts as an interface between perception, long-term memory and action (Baddeley 2003, 2012). This definition emphasizes WM as a top-down mental process, which has been shown to be crucial for higher-order cognitive functions such as problem solving, language comprehension, arithmetic and decision making (Diamond 2013). Thus, impairments of WM due to neurological, psychiatric and developmental disorder or as a result of aging have a large impact on work performances or daily functioning (Redick 2019; Glisky 2007). As a consequence, WM trainings were developed, aiming at increasing WM capacity (Katz, Shah, and Meyer 2017; Melby-Lervåg, Redick, and Hulme 2016).

Following the first promising results of WM training studies (Klingberg et al. 2005; Jaeggi et al. 2008) the field emerged and resulted in a large body of literature with the common aim of investigating the efficacy of various approaches to improve WM capacity. In experimental studies, the efficacy of WM trainings is typically investigated by the extent of *transfer effects*, i.e., if possible gains can be generalized to untrained cognitive tasks (Teixeira-Santos et al. 2019). Transfer effects can either occur as *near transfer*, which describes improvement on untrained tasks similar to the trained tasks (e.g. an untrained WM task) or as *far transfer*, describing improvement on a task of a different cognitive function (e.g. a fluid intelligence task) (Teixeira-Santos et al. 2019; Jaeggi et al. 2008). Although literature agrees on the efficacy of WM trainings in producing near transfer effects, far transfer effects remain a much-debated topic up to date. Indeed, meta-analytic reviews described near transfer effects to verbal and visuospatial WM tasks following WM trainings (Melby-Lervåg, Redick, and Hulme 2016; Soveri et al. 2017; Karbach and Verhaeghen 2014; Teixeira-Santos et al. 2019; Basak, Qin, and O'Connell 2020), which, moreover, maintained in older healthy adults in the long term (Teixeira-Santos et al. 2019). Although far transfer effects were reported following WM trainings (Soveri et al. 2017), they have been described to be small (Karbach and Verhaeghen 2014; Teixeira-Santos et al. 2019; Basak, Qin, and O'Connell 2020) and it has been concluded that there is no convincing evidence for far transfer effects (Melby-Lervåg, Redick, and Hulme 2016).

One main challenge in WM training research displays the diversity of training programs, which has been suggested to explain the controverse results of transfer effects (Pergher et al. 2020). So has type of training been described as a mediating factor of WM training efficacy (Teixeira-Santos et al. 2019; Morrison and Chein 2011). Indeed, various WM training approaches have been proposed over the last decades. Whereas *strategy-based trainings* aim at promoting specific strategies that facilitate the encoding, maintenance or retrieval of information, *multidomain trainings* lay their focus on training multiple cognitive functions next to WM with aiming at improving at least one of the trained functions (Morrison and Chein 2011; Karbach and Verhaeghen 2014). *Process-based* WM trainings, however, focus on training the core mechanisms of WM (Morrison and Chein 2011; Teixeira-Santos et al. 2019) and are usually computerized. Typically, they are either based on simple span tasks targeting the storage component of WM (Klingberg et al. 2005; Könen, Strobach, and Karbach 2016) or complex span tasks that target the updating, binding and processing function of WM (Redick and Lindsey 2013). While span trainings typically include multiple task, the “dual n-back training” (Jaeggi et al. 2008) is a widely used process based WM training assuming that visual and auditory WM are trained concurrently in one single task using auditory and visuospatial stimuli (Morrison and Chein 2011). However, it has been concluded that using different training tasks on multiple components of WM is more effective in producing near and far transfer gains compared to single-task trainings (Basak, Qin, and O’Connell 2020). The vast number of training paradigms may explain varying transfer effects, however hamper the possibility of comparing studies (Pergher et al. 2020). For this reason, recent WM training studies attempt to overcome this issue by investigating different training paradigms simultaneously to study their comparative efficacy. A direct comparison of a spatial n-back and a verbal complex span training showed near transfer from the n-back trainings to a new form of n-back task only, whereas no transfer was found for complex span trainings (Minear et al. 2016; Holmes et al. 2019). Furthermore, it has been described that n-back and complex span trainings do not only differ in their effectiveness on transfer but also in their underlying neural mechanisms (Blacker et al. 2017).

Additional factors have been identified to explain the controversial evidence of transfer effects. For instance, not all studies included active control groups undergoing a sham intervention in their design even though this has been suggested to be crucial in order to lead to evidence, that the WM training is causal for improvements in untrained tasks (Melby-Lervåg, Redick, and Hulme 2016). Further, the repeated administration of WM capacity tests to assess training gains has been shown to lead to practice effects (also refered to as retest or learning effects), which could overestimate the actual net training gains of WM trainings (Scharfen, Jansen, and Holling 2018). Although high practice effects have been described from the first to second test administration (Scharfen, Jansen, and Holling 2018), their control is mostly neglected in study designs. Moreover, the fact that most WM training programs were not based on theoretical models may have hampered optimal treatment effects and the understanding of the underlying mechanisms (Von Bastian and Oberauer 2013). Indeed, a WM training based on Baddeley’s WM model focusing on storage, selective attention and central executive processes was tested on its efficacy in a clinical trial in healthy old adults (Weicker et al. 2018). Each subprocess of WM was trained by a task to remember cards (storage module), focus on specific aspects of cards (selective attention module) and sort cards (manipulation module). The authors reported an increased WM performance on untrained tasks and an increase in everyday life activities, however, no far transfer was described, which was explained through the limited diverseness of the used training tasks (Weicker et al. 2018). Despite no far transfer was found, only a model-based structure of the training allows to draw conclusions on underlying processes that were trained and to identify task-related processes that may induce far transfer effects.

At last, task-related processes have been described to improve WM performance following training (Minear et al. 2016). Indeed, inhibitory abilities such as filtering efficiency – the ability to exclude irrelevant information from assessing WM (Li et al. 2017) - have been shown to render WM more efficiently than WM training alone (Schmicker et al. 2016). It has been described that older adults have difficulties to suppress task-irrelevant information during visual WM encoding and thus WM trainings are needed that train the exclusion of irrelevant information during WM encoding (Gazzaley et al. 2008; Jost et al. 2011). Indeed, a comparative study showed the same extent of improvement on WM tasks following filtering training on distractor inhibition compared to a WM training only (25). Nevertheless, only one study addressed far transfer following filtering efficiency training and did not find effects (Li et al. 2017). Additionally, although training of filtering efficiency has been associated with increased visual WM (Li et al. 2017), its effect on verbal WM has not been investigated yet. Thus, in order to understand if suppression of irrelevant information could display a task-related process for transfer effects following WM training, filtering efficiency should be embedded in verbal and visuospatial WM tasks. So far, no WM training study implemented filtering efficiency in WM tasks.

In summary, various approaches have been tested but no clear conclusion about their comparative efficacy can be drawn. They main cause of this uncertainty relates to the lack of theoretical background, differences in terms of experimental designs and the need of identifying task-related processes. In this parallel group randomized clinical trial we aimed at testing the efficacy of WM training based on Baddeley's multicomponent model (Baddeley, Hitch, and Bower 1974; Baddeley 2003; Baddeley, Allen, and Hitch 2011), which additionally trains filtering efficiency by 1) embedding it in the WM training and 2) targeting both verbal and visuospatial modalities. Whereas most WM trainings include tasks for training the phonological loop, visuospatial sketchpad and central executive, our training additionally includes a task for the episodic buffer resulting in a model-based WM training (MB). We implemented novel task levels that target filtering efficiency in the context of WM and added them to the MB training (MB^+). Both trainings (MB and MB^+) will be tested for their efficacy in improving WM performance by comparing them to a dual n-back training (Jaeggi et al. 2008) and an active control group. In order to minimize the learning effects related to the repetition of the assessment, a double baseline design is implemented as suggested previously (McCaffrey and Westervelt 1995; Duff et al. 2001). We hypothesize, that the MB^+ training shows superiority in improving WM performance and inducing transfer effect compared to a dual-n-back training and an active control group.

2 Materials and Methods

2.1 Participants

Based on our power calculations, we targeted to include a total of 120 subjects as the final sample with complete study termination, yielding 30 subjects in each of the four intervention groups to reach a moderate effect size (power = 0.8 and alpha = .05, two-sided). Subjects were recruited through online advertisement, advertisement in public transportation and courses for seniors at the University of Basel. Inclusion criteria were the presence of an informed consent as documented by signature and age 50 years old or older. Participants were excluded if they had 1) a medical history of psychiatric or neurological disorder assessed with a health status questionnaire and the Montgomery Åsperg Depression Rating Scale; MADRS (Montgomery and Asberg 1979), 2) a history of substance abuse, 3) a Benzodiazepine intake on a daily basis, 4) a colour vision deficiency defined by less than 13 correct answers at the Ishihara test (Ishihara 1987), 5) disability upper limbs that limits the use of tablet devices or 6) less than 26 points on the Montreal Cognitive Assessment (MoCA; (Nasreddine et al. 2005)). Written informed consent was obtained from each participant after a detailed explanation of the study procedures. The study was approved by the local ethics committee (Ethikkommission Nordwest und Zentralschweiz) and was conducted in accordance with the Declaration of Helsinki. Participants were reimbursed with CHF 100.- for their participation.

2.2 Experimental Design and Procedures

A longitudinal, parallel-group, randomized controlled trial design was employed. Although the study was designed as double blind and participants had no information about the performed training, it is not possible to fully blind the participants towards detecting which training group they were allocated to. For this reason the study is refered to as single-blind. After initial recruitment over the phone, participants were invited to our institute for a thirty-minute screening session where the eligibility criteria were verified for each participant and written informed consents were obtained. After inclusion in the study, all participants underwent four cognitive assessment sessions. The first two sessions (pre-baseline and baseline) took place within three weeks. At the second session (baseline), participants were randomly allocated to one of the four experimental groups using a minimization approach stratifying the sample regarding age and education. At baseline, participants received the tablet device with the corresponding training as well as a detailed explanation of it. To ensure the single blindness of the study, the study personnel were divided in training explanators, which explained the tablet and the training and provided a short example of each task to the subjects, and cognitive testers. Training explanators were not involved in any procedures addressing the cognitive testing. After the second session, participants were asked to train three weeks on the tablet devices at home. After three weeks and the completion of training program, participants underwent a third assessment session to capture possible training effects. An additional assessment session was performed at 12 weeks after the completion of training to investigate long-term effects. Each assessment session took place in a single subject setting and had a duration of approximately two hours (Fig. 1).

2.3 Trainings

The participants performed the trainings at home using touch-screen tablet devices provided by the research group. All trainings were implemented using Java in Android Studio v1.5.1 (https://developer.android.com/studio/index.html) and downloaded on Lenovo TAB A10 with Android 4.4 as operating system. All tasks were based on an adaptive design where difficulties of the tasks were modulated according the participants' performance. In order to guarantee a comparability of the trainings, the four training regimes were created by using the same visual design, length of the training session and number of sessions. All participants were instructed to train for three weeks, four sessions a week, 45 minutes each. In order to assess adherence to this training regimen the date and time of each training were logged and participants were additionally asked to note the date and time of completed training in a diary. Participants were allowed to choose the four days of training in one week, however, the training was programmed in a way that only one training session was possible each day. Participants were included in the final analysis upon completion of a minimum of 80% of the training (9 training sessions).

2.3.1 MB and MB$^+$ Training

Our in-house developed model-based WM trainings aimed at training participants on visuospatial and verbal WM as well as the central executive and episodic buffer function of WM on the theoretical ground of the multicomponent model (Alan Baddeley, Hitch, and Bower 1974).

Based on literature review, for each component – phonological loop, visuospatial sketchpad, episodic buffer and central executive - the task with the highest reliability and validity was chosen as the basis for the MB and MB$^+$. For both trainings, the assessment tasks were then transformed into a training task by creating adaptive levels of difficulty based on number of items and speed. For the MB$^+$, additional levels were created based on distractor inhibition corresponding to filtering efficiency for the visuospatial sketchpad and phonological loop tasks (Li et al. 2017; Baddeley 2012). Table 1 provides an overview over tasks and levels of the MB and MB$^+$ trainings. For the phonological loop, a simple letter span test was used as the basis of the training task. For the MB training, increasing item length was used as level of difficulty. In order to incorporate distractor inhibition for the MB$^+$ training, 1) presence of irrelevant background sounds on the ground of the "irrelevant noise effect" and 2) presentation of dissimilar or similar digits of items following the "similarity effect" (Baddeley 2003) was added to the increased sequence length as additional difficulty levels. For the visuospatial sketchpad, a visual pattern span test (Della Sala et al. 1999) was the basis for the visuospatial subcomponent. In this task, participants were asked to recall the pattern of colored squares in a grid by filling in the right positions in an empty grid, whereas difficulty increased with the size of the grid. For the MB$^+$ training, a level of difficulty was added by including an irrelevant visually loaded picture during the retention phase and the recall of the grid squares. A Corsi block tapping test (Baddeley 2003; Corsi 1972) formed the basis for the spatial subcomponent. In this task, participant had to recall flashing objects in the same order they appeared in the arrangement by tapping on the objects. The task improved in difficulty by increasing the item sequence. For the MB$^+$ training, a haptic irrelevant movement task was added between the presentation and recall of the object sequence task to the increasing sequence length of the presented items. The central executive component was targeted with a dual task in order to address the ability to focus, divide and switch attention (Mohr and Linden 2005; Logie et al. 2000). For the dual-task, participants were asked to complete a visual recall task and simultaneously a phonological task of recognizing high and low tones. Difficulty increased by increasing the speed and number of items. For the episodic buffer, we implemented a unimodal binding task where a spatial order of letters embedded in a frame are presented on a stimulus slide. Participants were asked to remember the exact position and letter and recall both in probe slides. Increased difficulty was achieved by adding letter/position combinations to the stimulus slide. The central executive and episodic buffer task were the same for both the MB and MB$^+$ training.

2.3.2 Dual n-back Training

The basis for the dual n-back training was the widely used "dual-n-back training paradigm" (Jaeggi et al. 2008). A complex dual n-back task including a visual and an auditory WM task was implemented according to the original publication, fit to tablet devices and designed to assure the comparability with the other trainings. We used our implemented graphic items instead of squares for the visuospatial task and letters in German language. Except for these adaptions, all parameters on item presentation and retention phases were kept as implemented in the original version.

2.3.3 Control Training

As a last comparator group, we included an active control intervention, since active control groups have been suggested to be more reliable in order to prove the specificity of WM training effects (Weicker, Villringer, and Thöne-Otto 2016) and have the function to be able to control for intervention and Hawthorne effects (von Bastian and Oberauer 2014). The control intervention in our study was as well developed in-house and consisted of three training tasks addressing manual dexterity, visual-motor coordination and fine motor control. For the manual dexterity task, subjects had to execute a series of finger movements following a visual cue and were asked to touch circles on the tablet screen that will change color with the corresponding finger. With increasing level, the speed of the presented visual cue and the number of cues increased. For the visual-motor coordination task, the participants are asked to follow the lines of presented letters which they heard also through headphones. With increasing difficulty, the letters were displayed incomplete and subject had to complete the presented letter which they heard by drawing them on the tablet. For the fine motor control task, the participants were asked to erase presented moving objects on the screen by executing a swishing movement using the index finger in a dedicated strip at the bottom of the screen. With increasing difficulty, the number of objects moved from the top of the screen to the bottom increased as well as the speed of the objects moving down.

2.3.4 Expectation Towards Improvement

The comparability of the four trainings and the expectations towards the improvement in the main outcomes of the different trainings were tested in a separate group of subjects not included in the trial. Volunteers aged around 50 years or older were ask to test the training for a few days and give feedback about their expectation for improving in WM tasks following the training. After a detailed explanation the training and the main outcome measures, we asked a total of 20 people (five for each training group) in the similar age range of the target sample (M = 60.58, SD = 10.35) on how much they would expect to improve on the main outcome after perceiving the training. They had to rate their improvement on a 10-point Likert scale from 0 = "I will not improve in this task by this training at all" to 10 = "I will improve in this task through this training very much". A Kruskal-Wallis Test was applied to investigate if the groups differ in their expectancy ratings. The results yielded no significant differences between the groups, indicating that participants had similar expectations towards the efficacy of the training and would therefore not detect the control training as such.

2.4 Cognitive Assessment

Cognitive assessment was performed at all four sessions in order to assess possible improvements on WM as well as near and far transfer. To cover a spectrum of transfer tasks, the assessment of cognitive functions included standardized tests (both pencil and paper as well as computerized tests) addressing WM, verbal and visual learning, processing speed and fluid intelligence. Parallel test forms were applied for tests investigating memory recall to further account for learning effects in the applied items. The assessments were performed at the Division of Cognitive Neuroscience by trained psychologists.

2.4.1 Working Memory

To investigate the near transfer of the trainings to WM, tasks were included that assess the storage, rehearsal and processing functions of WM. In old age it has been shown, that age-related effects in complex WM span tasks are higher than in single span tasks, since complex WM span tasks require the coordination of concurrent storage and processing, which is absent in single span tasks. Indeed, larger decreases in performance have been shown for complex WM span tasks than for single span tasks (Bopp and Verhaeghen 2005). In addition, it has been stated that a more 'pure' measure of WM capacity can be derived from using three complex span tasks than from only using one measure to assess WM (Conway et al. 2005). For these reasons, WM transfer as a main outcome was assessed using the shortened versions of the Rotation Span Task, Symmetry Span Task and the Operation Span Task (Foster et al. 2014). It has further been shown, that the complex WM span tasks measure a domain-general capacity of WM and highly correlate with each other even though the altering content of the single task (Foster et al. 2014). For this reason, already in previous studies complex WM tasks were translated into a composite score (Borella and Carretti 2008; Chiaravalloti, Genova, and DeLuca 2015). In our study, we similarly created a composite score out of the operation, rotation and symmetry span tasks by building a sum of the partial score of each task, which then was used as the main outcome in the statistical model.

2.4.2 Near Transfer

In order to exploratory investigate, if possible training gains are limited to the underlying tasks of the model-based trainings, we included the Corsi Block Tapping Test which assesses Visuospatial WM (Corsi 1972) and as well as a visuospatial n-back task (Bürki et al. 2014) and the Forward and Backward Digit Span (WAIS-IV (Wechsler 2008)) task which assesses auditory WM. Near transfer to the WM related cognitive functions verbal learning were assessed using the Rey Auditory Verbal Learning Test (RAVLT (Schmidt 1996)), to recall of nonverbal information the Rey Osterrieth Complex Figure Test (ROCFT (Rey 1941; Osterrieth 1944)) and to executive functions were measured using the Trail Making Test form A and B (Reitan 1958).

2.4.3 Far Transfer

Reasoning abilities were assessed using Raven's Standard Progressive Matrices (SPM (Raven and Court 1996)). In order to reduce the administration time, a 9-item short form of the SPM has been crated and extensively tested on its psychological property in healthy controls and patients with schizophrenia. Results revealed a correlation of r = .9836 (Form A) and r = .9782 (Form B) with the original 60-Item form of the SPM which let the authors conclude, that the properties of the short forms are comparable to the original form of the SPM (Bilker et al. 2012). As a last secondary outcome measures, the Depression, Anxiety and Stress Scale (DASS-42 (Lovibond and Lovibond 1995)) was used to assess depression, anxiety and stress symptoms.

2.5 Statistical Analysis

Demographic factors were compared among groups using ANOVAs and chi-square test. Baseline differences were analysed using an ANOVA model with baseline performance as outcome and group as the between group factor. In order to investigate possible repetition effects between the pre-baseline and baseline session, a linear mixed-effects model was carried out on the whole group level with session as the within-subjects factor on test performance on all tasks.

In order to analyse the training gains in all outcome tasks, pre- and post-training session of the outcome measures were analysed using a linear mixed-effects model with the interaction term session x training group as fixed effect and study participant as random effect. An advantage of applying linear mixed-effects models in the analyses of longitudinal data is the ability to account for missing data points (Krueger and Tian 2004). For this reason, subjects were also included in the analyses if they did not complete all sessions due to the early termination of the study. In order to investigate the superiority to other training approaches, the MB^+ training was set as baseline comparator using an *a priori* contrast. *F* statistics were gained by running an ANOVA over the linear model using type II sums of squares. The same statistical analyses were applied for the long-term gain analyses between the post-training and follow up session at three months. In order to investigate differences in means pre- and post-training and post-training to follow up, univariate analyses within the groups were done on all complete cases using paired *t*-tests. For each statistical analysis, a p-value < 0.05 was considered as significant. All data were analyzed in R Studio, Version 1.2.1335 (R Core Team 2013).

3 Results

Due to the COVID-19 pandemic and subsequently unplanned early determination of the study the aimed sample size could not be reached. Between July 02, 2019 and the early termination on March 13, 2020, 161 participants from the German speaking part of Switzerland were screened for participation, of whom in total 38 either did not meet the eligibility criteria, withdrew interest before the study start or could not start due to the COVID19 outbreak. 123 healthy subjects aged from 50-81 (78 women and 45 men; aged 64.1±8.3 years) were included in the study and completed the pre-baseline session. 109 (68 women and 41 men; aged 64.5±8.2 years) completed the pre-baseline and the baseline session. 93 subjects (59 women and 34 men; mean age: 64.3±7.8 years) completed the pre-baseline, baseline and post-training session as well as the training period. Out of this sample, 60 subjects completed the whole study participation (42 women and 18 men; mean age: 64.2±7.2 years). Figure 2 shows the trial profile and included sample. Demographics of the included sample at pre-baseline are listed in Table 2. All 123 subjects were randomized to one of the four training groups after the pre-baseline yielding an n = 29 in the MB^+, n = 32 in the MB and n = 33 in the dual n-back training group and n = 29 in the control intervention group. All groups did not differ ($p < .05$) in age, years of education, MoCA, MADRS and Ishihara score at pre-baseline.

3.1 Training Data

93 participants received a tablet for training and were instructed to train four times a week for 3 weeks. Two participants were excluded for the analyses of pre- and post-training performance since one participant completed three and another one 24 training sessions. In average,

participants completed 11.82 ± 0.81 training sessions. 75 out of 92 subjects (81.5%) completed 12 training sessions, however, 90 subjects (97.8%) of the included 92 participants completed the for the inclusion required 80% percent of the training (9 or more training sessions).

3.2 Analyses of Baseline Performance at Whole Group Level

Results indicated no difference in baseline performance for the complex span score between all groups in neither the pre-baseline (($F(3, 115) = 1.60$, ns) nor the baseline session (($F(3, 102) = 1.04$, ns) for the complex span composite score.

Regarding the near transfer tasks, no difference between all groups in baseline performance neither at the pre-baseline nor baseline were present. For the other outcomes of interest, only the digit span task showed a significant group difference between groups at the pre-baseline ($F(3, 119) = 3.52$, $p < 0.05$), however the group difference was not present at the baseline session. For the corsi block tapping test, and the n-back test groups did not differ in their performance at pre-baseline or baseline.

3.3 Analyses of Practice Effects Between Double-baseline at Whole Group Level

A linear mixed-effects model comparing the pre-baseline to the baseline performance in the complex span composite score revealed a significant effect for session ($\chi^2(1) = 16.20$, $p < .001$), indicating a repetition effect. On the whole group level, there was significant increase in performance on the complex span composite score at baseline compared to pre-baseline ($b = 4.85$, $t(103) = 4.01$, $p < .001$, $r = 0.37$), despite no training took place between these sessions.

Regarding the transfer tasks, there were significant effects for session in the 30 min recall of the ROCFT ($\chi^2(1) = 19.4$, $p < .001$) with a significant increase in performance at baseline compared to pre-baseline ($b = 2.07$, $t(108) = 4.38$, $p < .001$, $r = 0.39$), the TMT Form B ($\chi^2(1) = 18.37$, $p < .001$) with a significant decrease in reaction time at baseline compared to pre-baseline ($b = -9.61$, $t(108) = -4.27$, $p < .001$, $r = 0.38$), the n-back task ($\chi^2(1) = 10.51$, $p < .01$) with a significant decrease in wrong answers at baseline compared to pre-baseline ($b = -2.92$, $t(106) = -3.23$, $p < .001$, $r = 0.30$), the DASS anxiety subscale ($\chi^2(1) = 4.80$, $p < .05$) with a significant decrease in score at baseline compared to pre-baseline ($b = -0.37$, $t(106) = -2.18$, $p < .05$, $r = 0.21$), the DASS depression subscale ($\chi^2(1) = 5.59$, $p < .05$) with a significant decrease in score at baseline compared to pre-baseline ($b = -0.55$, $t(106) = -2.35$, $p < .05$, $r = 0.22$) and the DASS stress subscale ($\chi^2(1) = 4.52$, $p < .05$) with a significant decrease in score at baseline compared to pre-baseline ($b = -0.79$, $t(106) = -2.12$, $p < .05$, $r = 0.20$). No changes between the two assessment sessions were found for the RAVLT, the SPM, the Corsi test and the digit span test. All means and standard deviations of the pre-baseline and baseline session are displayed in table 3.

3.4 Training Effects on the Complex Span Composite Score

The linear mixed-effects model revealed a significant main effect for session ($\chi^2(1) = 10.56$, $p = .001$) and a tendency for significance in the interaction group x session ($\chi^2(3) = 7.29$, $p = .063$) for the composite score between baseline and post-training. Setting the MB^+ group as an a priori contrast showed, that there was a significant difference post-training between the MB^+ and the MB training ($b = -9.62$, $t(82) = -2.52$), $p = .014$, $r = 0.27$) and a tendency for significance in the comparison between the MB+ and the dual-n-back training ($b = -7.59$, $t(82) = -1.87$), $p = .065$, $r = 0.20$) and the control intervention ($b = -7.08$, $t(82) = -1.86$), $p = .067$, $r = 0.20$). From post-training to the follow-up at 3 months, the linear mixed effects model showed no significant effects for group ($\chi^2(3) = 1.56$, p = ns), session ($\chi^2(1) = 0.40$, p = ns) or the interaction between group and session ($\chi^2(3) = 2.99$, p = ns) for the complex span composite score.

Results of univariate analyses showed a significant within group difference between the pre- and post-training session in the MB^+ training group ($t(22) = -3.34$, $p < 0.05$). All other trainings and the sham interventions showed no differences in pre- and post-training on the complex span composite score in univariate analyses. Results show no significant group effect for the difference of the complex span composite score between post-training and follow up after 3 months. As well the univariate analyses showed no significant differences in the composite score between the post-training and follow up in the MB^+, MB and dual-n-back training. However, the univariate analyses of the control training showed a significant decrease in performance on the composite score from the post-training session to the follow up ($t(13) = 2.56$, $p < .05$). Mean, standard deviation and effect sizes for the univariate within group analyses are reported in table 4.

3.5 Analyses of Near, Far Transfer and Additional Outcomes

The linear mixed-effects models applied to investigate the training gains between groups for all transfer tasks showed a significant interaction effect for the TMT form B ($\chi^2(1) = 8.06$, $p = .04$). The contrast however revealed no significant difference between any of the groups with the MB^+ training. All other models yielded no significant interaction effects for near or far transfer measures pre- to post-training. There was a significant effect for session in the RAVLT ($\chi^2(1) = 4.89$, $p = .03$), the ROCFT ($\chi^2(1) = 6.54$, $p = .01$) and the n-back task ($\chi^2(1) = 19.27$, $p < .001$) in the linear mixed-effects models testing group differences to the MB^+ training. Additionally, a significant group effect was present in the n-back task ($\chi^2(1) = 8.4$, $p = .04$). For all other tasks, neither group nor session main effects were present.

Regarding the post-training to follow up, the linear mixed-effects model showed a significant interaction between group and session for the DASS stress subscale ($\chi^2(3) = 19.27$, $p < .001$). The investigation of the contrasts showed a significant difference at the follow up session between the control group ($b = 2.88$, $t(51) = 2.54$), $p = .014$, $r = .33$) as well as the dual-n-back training ($b = 2.49$, $t(51) = 2.28$), $p = .027$, $r = .30$) and the MB^+ training. Furthermore, there was a significant main effect for session in the TMT form B ($\chi^2(1) = 4.43$, $p = .04$) and a significant effect for group in the n-back task ($\chi^2(1) = 8.98$, $p = .03$). All other outcome measures showed no effects for the group, session and interactions between group and session. All linear mixed-effects model outcomes for the interaction effects are reported in table 5.

The univariate analyses within the MB^+ group showed a significant improvement on the ROCFT 30 min recall post-training ($t(21) = -3.8$, $p < .05$), indicating a transfer effect to this task. All other within group comparisons showed no significant changes, neither at pre-training to post-training nor comparing post-training and follow up. The within group analyses of the MB training showed a significant decrease in reaction time post-training in the TMT form B ($t(23) = 2.19$, $p < .05$) and a significant decrease in wrong answers at the n-back test post-training ($t(23) = 2.12$, $p < .05$). There were no other training related changes or changes at the follow up session. For the dual-n-back training, a significant decrease in wrong answers in the n-back task were found post training ($t(25) = 3.83$, $p < .001$). There were no other changes in the transfer tasks comparing pre- and post-training. There were also no changes between the post-training and follow up session. Within group analyses of the control training showed no training related changes comparing pre- and post-training. However, there was a significant increase in the DASS depression ($t(13) = 2.19$, $p < .05$) and the DASS anxiety subscale ($t(13) = -2.75$, $p < .05$) at the 3-moth follow up. There were no other training-related changes or changes between the post-training and follow up session.

4 Discussion

In this parallel group randomized clinical trial we investigated two computerized WM trainings based on the multicomponent model (A. Baddeley, Hitch, and Bower 1974) with (MB^+) and without (MB) inclusion of distractor inhibition on their efficacy of improving WM performance and inducing transfer effects. Both trainings were compared to a dual n-back training and an active control intervention in healthy old adults. After accounting for practice effects, only the MB^+ training group shows an improvement in WM capacity tasks. Compared to a model-based, a dual-n-back and a control training, the MB^+ training shows an overall tendency for superiority in improving WM capacity, which was particularly evident compared to the MB training. The dual-n-back, MB and control group showed no improvements on WM capacity. Regarding transfer to trained and untrained cognitive functions, the MB^+ group showed an improvement in visuospatial learning, the MB group an improvement in a processing speed and visuospatial n-back task and the dual-n-back group an improvement in an untrained visuospatial n-back task. In the direct comparison among trainings, transfer effects were only detected to the processing speed and a visuospatial n-back task following the MB training. From post-training to the 12-week follow up, the control training group showed a decrease in performance on WM capacity tasks.

Our results show that although two trainings were developed on the basis of the same theoretical model, only the MB^+ group, which include the distractor inhibition component improved the WM capacity. While the effect was only tendentially significant in the overall comparison between all trainings, this tendency is supported by the univariate analyses, which showed improvement on untrained WM capacity tasks only in the MB^+ group, even after accounting for practice effects. This finding is in line with previous studies describing improvements on WM tasks following training on filtering efficacy (Shin et al. 2015; Li et al. 2017; Schmicker et al. 2016). In comparison to previous studies that investigated filtering training and WM training separately, our results expand those findings in the sense that distractor inhibition was embedded in the WM training tasks. In addition, we found a near transfer effect to visuospatial learning following the MB^+ training. Although a previous study on effects of filtering efficiency training did not find transfer effects to other cognitive functions, improvement specifically on visuospatial WM were described (Li et al. 2017). Since the improvement on visuospatial learning in our study was not present in the comparison to other trainings, further studies are needed in order to investigate far transfer effects of filtering in combination with WM training. Nevertheless, following our results and those of previous studies, the question arises, if filtering efficiency could constitute a task-related process of WM training in old age. Indeed, it has been suggested that filtering training seems to benefit WM improvement by increasing selection abilities which could in turn increase the efficiency of memory encoding (Schmicker et al. 2016). Additionally, it has been suggested that older adults use distractors in cognitive tasks as environmental support in order to counterbalance decreasing cognitive performance (Rumpf et al. 2019). In light of the previous suggested association between reduced WM capacity and reduced filtering efficiency in old age (Jost et al. 2011), training distractor inhibition in the context of a WM as implemented by the MB^+ training could therefore enhance the selection ability during WM tasks and by that facilitate the completion of a complex WM task, which in turn enhances WM capacity.

This mechanism of action could also explain the absence of improvement on WM capacity following the MB training, which did not train distractor inhibition. In the direct comparison of the training approaches, differences between the trainings in regard to the transfer effect were only found for the TMT form B, indicating improved processing speed following the MB training. This effect is also supported by the univariate analyses, where improvement on the TMT form B and additionally small improvement on the visuospatial n-back task was found. Although the TMT is designed to measure processing speed, it has been described that a simple span task explained most variance of the TMT form B, indicating a reflection of WM (Sánchez-Cubillo et al. 2009). Additionally, small effects on improvements on simple span tasks following n-back training have been reported previously (Soveri et al. 2017). It could therefore be assumed, that the MB training with its tasks structured according to simple span affects WM-related tasks, however only a combination of the MB-tasks with distractor inhibition as implemented in the MB^+ training has the ability to tap WM capacity.

Still, in order to understand if the combination of distractor and WM training indeed displays a task-related process, future studies should investigate this combination of filtering and WM training compared to filtering training or WM training alone in order to understand its benefits for WM capacity in old age.

Besides the differences between the two model-based trainings, we investigated the efficacy of the MB$^+$ training compared to a dual-n-back. Our results showed a tendency for difference between the MB$^+$ and the dual-n-back training in regard to improved WM capacity, which is supported by the univariate analyses indicating an absence of training gains on WM capacity in the dual-n-back training. Additionally, improvement on an untrained visuospatial n-back task was found following the dual-n-back training. This finding is in line with a meta-analysis which investigated the efficacy of 33 studies and described a moderate effect of transfer to untrained n-back tasks and a small effect to other untrained WM tasks, concluding that the transfer effects following n-back training remain task-specific (Soveri et al. 2017). Likewise, a comparison between a spatial n-back and a verbal complex span training showed no training gains to untrained complex span tasks but an improved performance on an untrained n-back task following the spatial n-back training (Minear et al. 2016). Holmes et al. (2019) correspondingly described an improvement on an untrained n-back task but no cross-paradigm transfer to a verbal complex-span task following n-back training in a direct comparison of both training paradigms. Additionally, a comparison between n-back and arithmetic updating training and their effects on updating and complex WM task yielded improvements only in outcome tasks that were structurally similar to the trained function (Linares et al. 2019). Our results therefore support previous studies which concluded, that transfer from n-back trainings in comparison to other training strategies is task-specific and extent those findings by drawing this conclusion in a sample of old adults.

Next to achieving improvements on untrained WM tasks, producing long-term and far transfer effects to other cognitive functions is of main interest. Our results suggest no far transfer from neither model-based (MB and MB$^+$) nor dual-n-back training to reasoning measured by the SPM test. This is in line with previous research, which reported no far transfer to reasoning following WM training (G. Sala et al. 2019; Teixeira-Santos et al. 2019; Minear et al. 2016) and filtering training (Li et al. 2017). The lack of far transfer effects to reasoning performances following WM trainings may question the importance of WM training in the old population (G. Sala et al. 2019). However, amongst other factors that could explain the absent far transfer effects, the type of outcome measure has been suggested as a moderator, since slightly higher transfer effects were described in reasoning abilities measured with the Cattell test versus the SPM test (Teixeira-Santos et al. 2019). The authors suggest, that measures such as the Cattell test with its division in subtests may reflect reasoning more comprehensive and therefore highlight the importance of used measurement to assess training gains. Additionally, a recent study investigated the far transfer effect of WM training in old age by measuring everyday functioning, and reported improvements not only at post-test but also at a 6-month follow-up (Erika Borella et al. 2019). These findings indicate, that transfer effects can occur and highlights the importance of shifting the focus from investigating improvements on other cognitive tasks towards transfer to daily life activities in future studies.

At last, three methodological aspects in the field of WM training studies should be discussed. First, although practice effects have been described in in WM capacity tasks (Scharfen, Jansen, and Holling 2018), most WM training studies do not account for them in their study designs. Our results suggest substantial practice effects in complex span tasks as well as a visuospatial n-back task after a repeated test administration before training. This is in line with a meta-analysis, which described practice effects in WM tasks and specifically reported larger practice effects in updating, n-back, complex span and coordination tasks than for simple span tasks, concluding that unfamiliar and challenging cognitive tasks are more subject to practice effects (Scharfen, Jansen, and Holling 2018). As one explanation for the occurrence of practice effects, interference of anxiety has been described, which has been found to be reduced largest after a second and reaching a plateau after a fourth administration of cognitive testing (Jendryczko, Scharfen, and Holling 2019). In our sample, we found reduced scores on all three subscales of the anxiety, stress and depression scale at the second test administration and no changes in all three scores between pre- and post-training. Therefore, our results support previous findings suggesting that complex span and n-back tasks may be perceived as difficult by the participants and may induce stress and anxiety, which could further also be related to the unfamiliarity of the testing situation. Following our results, we suggest that perceived stress and anxiety may be reduced by a second test administration before the WM training and should therefore be taken into consideration in form of a double-baseline design in WM training studies. This approach could account for practice effects and hence help in detecting the true training gains following WM trainings.

Second, the selection of appropriate control condition displays an issue in WM training studies (Morrison and Chein 2011; Shawn Green et al. 2019). It has been recommended, that active control condition should be used in training studies in order to account for various effects such as familiarity of testing situation and motivation towards training (Melby-Lervåg, Redick, and Hulme 2016; Morrison and Chein 2011), however, that they are not properly controlling for expectancy effects (Boot et al. 2013; Morrison and Chein 2011). In our study the control condition consisted of a sham intervention with a similar design of the training conditions, performed on the tablet device. A pilot investigation in a separate sample before the clinical trial yielded no differences between the trainings related to expectancy, indicating that the participants had similar expectations towards their improvements. Nevertheless, it has been suggested that mechanistic studies – whose goal is to identify underlying mechanisms – should additionally include a passive control group, which could help in the interpretation of absent differences (Shawn Green et al. 2019). In our study we did not include a passive control group. Nevertheless, our study design allowed conclusions regarding the mechanism of action, since only distractor inhibition was manipulated in one of the two model-based trainings. Future studies should carefully choose the appropriate control group(s) in order to gain insight in the trained mechanisms.

Third, the variability of assessment tests, training tasks, paradigms and transfer measures has been suggested to be a severe issue in order to draw conclusions across studies (Pergher et al. 2020). Although comparative studies – such as ours - help to counteract this issue, future studies should find a consensus for the assessment of transfer by using valid and appropriate tasks, which further investigate the application of the training in daily life (Pergher et al. 2020; Erika Borella et al. 2019).

Despite the vast control of methodological issues and theoretical considerations applied in this clinical trial, we have to acknowledge several limitations. First - and most important –, the *a priori* calculated sample size could not be reached due to the early termination of the study because of restrictive measures due to the COVID-19 pandemic. For this reason, the study is underpowered and conclusion have to be interpreted with caution. This is specifically evident in the interpretation of the long-term transfer measured at the 3-month follow-up. On the univariate level, we could interpret that all training groups remained on their levels except for the control group, that showed a decrease on WM capacity at 3-months follow up, indicating long-term effects on WM capacity improvement in the MB^+ training. However, only half of the sample reached the follow up session and therefore this effect has to be interpreted with caution. Second, although the univariate analyses showed no improvements on WM capacity, the low power of the study due to the incomplete sample size could additionally account for the lack of significant difference between the active control group and MB^+ training in the comparison of all trainings. The uncomplete recruitment led to unbalanced group size and therefore limits the conclusions that can be drawn from the described effects.

In conclusion, our study suggests that a model-based WM training in combination with distractor inhibition in the sense of filtering efficacy is a promising approach to induce improvements in WM capacity. Although the study is underpowered, it shows that a rigorous methodological control by accounting for practice effects and choice of adequate control condition can lead to insights on the effects of possible confounding factors. Future studies are needed to investigate the described mechanism of action in large scale comparative studies and their far transfer effects to activities of daily life. In this way, we can develop efficient training programs and study their transfer effects, whether it is for the enhancement of cognitive functions and their applicability in everyday life or as a basis for effective rehabilitation programs.

5 Conflict of Interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest. Hoffmann-La Roche Ltd. was not involved in the research.

6 Author Contributions

PZ, DQ and SM designed the research. EG and PZ conducted the study. PZ performed the statistical analyses and worte the manuscript. All authors read and approved the final version of the manuscript.

7 Funding

This project was funded by the Swiss National Science Foundation (SNSF, Doc.CH P0BSP1_181741 to P.Z.) and received support from the Transfaculty Research Platform of the University of Basel.

8 Acknowledgment

The authors want to thank MSc Jan Bitterli, MSc Lauriane Arnegger, MSc Vera Nina Looser, BSc Martina Foresti, BSc Gresa Mazreku, MSc Galya Iseli, MSc Julian Gygax and MSc Melike Sahin for their contribution in the data collection and development of the trainings, and all participants for their participation in the study. The study was supported by the Swiss National Science Foundation (SNSF; Doc.CH P0BSP1_181741 to P.Z.).

Baddeley, Alan. 2003. “Working Memory: Looking Back and Looking Forward.” *Nature Reviews Neuroscience* 4 (10): 829–39.

———. 2012. “Working Memory: Theories, Models, and Controversies.” *Annual Review of Psychology* 63 (1): 1–29. https://doi.org/10.1146/annurev-psych-120710-100422.

Baddeley, Alan D., Richard J. Allen, and Graham J. Hitch. 2011. “Binding in Visual Working Memory: The Role of the Episodic Buffer.” *Neuropsychologia* 49 (6): 1393–1400. https://doi.org/10.1016/j.neuropsychologia.2010.12.042.

Baddeley, Alan, Graham Hitch, and Gordon H Bower. 1974. “Working Memory. The Psychology of Learning and Motivation,” 47–89.

Basak, Chandramallika, Shuo Qin, and Margaret A. O’Connell. 2020. “Differential Effects of Cognitive Training Modules in Healthy Aging and Mild Cognitive Impairment: A Comprehensive Meta-Analysis of Randomized Controlled Trials.” *Psychology and Aging* 35 (2): 220–49. https://doi.org/10.1037/pag0000442.

Bastian, Claudia C. von, and Klaus Oberauer. 2014. “Effects and Mechanisms of Working Memory Training: A Review.” *Psychological Research* 78 (6): 803–20. https://doi.org/10.1007/s00426-013-0524-6.

Bastian, Claudia C. Von, and Klaus Oberauer. 2013. “Distinct Transfer Effects of Training Different Facets of Working Memory Capacity.” *Journal of Memory and Language* 69 (1): 36–58. https://doi.org/10.1016/j.jml.2013.02.002.

Bilker, Warren B., John A. Hansen, Colleen M. Brensinger, Jan Richard, Raquel E. Gur, and Ruben C. Gur. 2012. “Development of Abbreviated Nine-Item Forms of the Raven’s Standard Progressive Matrices Test.” *Assessment* 19 (3): 354–69.

Blacker, Kara J., Serban Negoita, Joshua B. Ewen, and Susan M. Courtney. 2017. “N-Back Versus Complex Span Working Memory Training.” *Journal of Cognitive Enhancement* 1 (4): 434–54. https://doi.org/10.1007/s41465-017-0044-1.

Boot, Walter R., Daniel J. Simons, Cary Stothart, and Cassie Stutts. 2013. “The Pervasive Problem With Placebos in Psychology: Why Active Control Groups Are Not Sufficient to Rule Out Placebo Effects.” *Perspectives on Psychological Science* 8 (4): 445–54. https://doi.org/10.1177/1745691613491271.

Bopp, Kara L., and Paul Verhaeghen. 2005. “Aging and Verbal Memory Span: A Meta-Analysis.” *Journals of Gerontology - Series B Psychological Sciences and Social Sciences* 60 (5). https://doi.org/10.1093/geronb/60.5.P223.

Borella, E, and B Carretti. 2008. “Working Memory and Inhibition across the Adult Life-

Span." *Acta Psychologica* 128 (1): 33–44.

Borella, Erika, Alessandra Cantarella, Barbara Carretti, Annalisa De Lucia, and Rossana De Beni. 2019. "Improving Everyday Functioning in the Old-Old with Working Memory Training." *American Journal of Geriatric Psychiatry* 27 (9): 975–83. https://doi.org/10.1016/j.jagp.2019.01.210.

Bürki, Céline N., Catherine Ludwig, Christian Chicherio, and Anik de Ribaupierre. 2014. "Individual Differences in Cognitive Plasticity: An Investigation of Training Curves in Younger and Older Adults." *Psychological Research* 78 (6): 821–35. https://doi.org/10.1007/s00426-014-0559-3.

Chiaravalloti, Nancy D., Helen M. Genova, and John DeLuca. 2015. "Cognitive Rehabilitation in Multiple Sclerosis: The Role of Plasticity." *Frontiers in Neurology* 6: 1–10. https://doi.org/10.3389/fneur.2015.00067.

Conway, Andrew R. a., Michael J. Kane, Michael F. Bunting, D. Zach Hambrick, Oliver Wilhelm, and Randall W. Engle. 2005. "Working Memory Span Tasks: A Methodological Review and User's Guide." *Psychon Bull Rev* 12 (5): 769–86. https://doi.org/10.3758/BF03196772.

Corsi, P. 1972. *Memory and the Medial Temporal Region of the Brain. McGill University*. Montreal, QB.

Diamond, Adele. 2013. "Executive Functions." *Annual Review of Psychology* 64: 135–68.

Duff, Kevin, Holly James Westervelt, Robert J. McCaffrey, and Richard F Haase. 2001. "Practice Effects, Test-Retest Stability, and Dual Baseline Assessments with the California Verbal Learning Test in an HIV Sample." *Archives of Clinical Neuropsychology* 16 (5): 461–76.

Foster, Jeffrey L., Zach Shipstead, Tyler L. Harrison, Kenny L. Hicks, Thomas S. Redick, and Randall W. Engle. 2014. "Shortened Complex Span Tasks Can Reliably Measure Working Memory Capacity." *Memory and Cognition* 43 (2): 226–36. https://doi.org/10.3758/s13421-014-0461-7.

Gazzaley, Adam, Wesley Clapp, Jon Kelley, Kevin McEvoy, Robert T. Knight, and Mark D'Esposito. 2008. "Age-Related Top-down Suppression Deficit in the Early Stages of Cortical Visual Memory Processing." *Proceedings of the National Academy of Sciences of the United States of America* 105 (35): 13122–26. https://doi.org/10.1073/pnas.0806074105.

Glisky, Elizabeth. 2007. "Changes in Cognitive Function in Human Aging." In

Books.Google.Com, 3–20. https://doi.org/10.1201/9781420005523.sec1.

Holmes, Joni, Francesca Woolgar, Adam Hampshire, and Susan E. Gathercole. 2019. “Are Working Memory Training Effects Paradigm-Specific?” *Frontiers in Psychology* 10. https://doi.org/10.3389/fpsyg.2019.01103.

Ishihara, Shinobu. 1987. *Tests for Colour-Blindness*. Tokyo: Kanehara Shuppan Co., Ltd.

Jaeggi, SM, M Buschkuehl, J Jonides, and W. J. Perrig. 2008. “Improving Fluid Intelligence with Training on Working Memory.” *Proceedings of the National Academy of Sciences of the United States of America* 105 (19): 6829–33.

Jendryczko, David, Jana Scharfen, and Heinz Holling. 2019. “The Impact of Situational Test Anxiety on Retest Effects in Cognitive Ability Testing: A Structural Equation Modeling Approach.” *Journal of Intelligence* 7 (4).

Jost, Kerstin, Richard L. Bryck, Edward K. Vogel, and Ulrich Mayr. 2011. “Are Old Adults Just like Low Working Memory Young Adults? Filtering Efficiency and Age Differences in Visual Working Memory.” *Cerebral Cortex* 21 (5): 1147–54. https://doi.org/10.1093/cercor/bhq185.

Karbach, Julia, and Paul Verhaeghen. 2014. “Making Working Memory Work: A Meta-Analysis of Executive-Control and Working Memory Training in Older Adults.” *Psychological Science* 25 (11): 2027–37. https://doi.org/10.1177/0956797614548725.

Katz, Benjamin, Priti Shah, and David E Meyer. 2017. “How to Play 20 Questions with Nature and Lose: Reflections on 100 Years of Brain-Training Research.” *National Acad Sciences*. https://doi.org/10.1073/pnas.1617102114.

Klingberg, Torkel, Elisabeth Fernell, Pernille J. Olesen, Mats Johnson, Per Gustafsson, Kerstin Dahlström, Christopher G. Gillberg, Hans Forssberg, and Helena Westerberg. 2005. “Computerized Training of Working Memory in Children with ADHD - A Randomized, Controlled Trial.” *Journal of the American Academy of Child and Adolescent Psychiatry* 44 (2): 177–86. https://doi.org/10.1097/00004583-200502000-00010.

Könen, Tanja, Tilo Strobach, and Julia Karbach. 2016. “Working Memory.” In *Cognitive Training: An Overview of Features and Applications*, 59–68. Springer International Publishing. https://doi.org/10.1007/978-3-319-42662-4_6.

Krueger, Charlene, and Lili Tian. 2004. “A Comparison of the General Linear Mixed Model and Repeated Measures ANOVA Using a Dataset with Multiple Missing Data Points.” *Biological Research for Nursing* 6 (2): 151–57.

https://doi.org/10.1177/1099800404267682.

Li, Cui Hong, Xu He, Yu Juan Wang, Zhe Hu, and Chun Yan Guo. 2017. “Visual Working Memory Capacity Can Be Increased by Training on Distractor Filtering Efficiency.” *Frontiers in Psychology* 8. https://doi.org/10.3389/fpsyg.2017.00196.

Linares, Rocío, Erika Borella, Mª Teresa Lechuga, Barbara Carretti, and Santiago Pelegrina. 2019. “Nearest Transfer Effects of Working Memory Training: A Comparison of Two Programs Focused on Working Memory Updating.” Edited by David Giofrè. *PLOS ONE* 14 (2). https://doi.org/10.1371/journal.pone.0211321.

Logie, Robert H., Sergio Della Sala, Val Wynn, and Alan D. Baddeley. 2000. “Visual Similarity Effects in Immediate Verbal Serial Recall.” *Quarterly Journal of Experimental Psychology Section A: Human Experimental Psychology* 53 (3): 626–46. https://doi.org/10.1080/713755916.

Lovibond, P. F., and S H Lovibond. 1995. “The Structure of Negative Emotional States: Comparison of the Depression Anxiety Stress Scales (DASS) with the Beck Depression and Anxiety Inventories.” *Behaviour Research and Therapy* 33 (3): 335–43. https://doi.org/10.1016/0005-7967(94)00075-U.

McCaffrey, Robert J., and Holly James Westervelt. 1995. “Issues Associated with Repeated Neuropsychological Assessments.” *Neuropsychology Review* 5 (3): 203–21. https://doi.org/10.1007/BF02214762.

Melby-Lervåg, Monica, Thomas S Redick, and Charles Hulme. 2016. “Working Memory Training Does Not Improve Performance on Measures of Intelligence or Other Measures of ‘Far Transfer’: Evidence From a Meta-Analytic Review.” *Perspectives on Psychological Science* 11 (4): 512–34. https://doi.org/10.1177/1745691616635612.

Minear, Meredith, Faith Brasher, Claudia Brandt Guerrero, Mandy Brasher, Andrew Moore, and Joshua Sukeena. 2016. “A Simultaneous Examination of Two Forms of Working Memory Training: Evidence for near Transfer Only.” *Memory and Cognition* 44 (7): 1014–37. https://doi.org/10.3758/s13421-016-0616-9.

Mohr, Harald M., and David E.J. Linden. 2005. “Separation of the Systems for Color and Spatial Manipulation in Working Memory Revealed by a Dual-Task Procedure.” *Journal of Cognitive Neuroscience* 17 (2): 355–66. https://doi.org/10.1162/0898929053124929.

Montgomery, S. A., and M. Asberg. 1979. “A New Depression Scale Designed to Be Sensitive to Change.” *British Journal of Psychiatry* 134 (4): 382–89.

Morrison, Alexandra B., and Jason M. Chein. 2011. “Does Working Memory Training Work? The Promise and Challenges of Enhancing Cognition by Training Working Memory.” *Psychonomic Bulletin and Review* 18 (1): 46–60. https://doi.org/10.3758/s13423-010-0034-0.

Nasreddine, Ziad S., Natalie A. Phillips, Valérie Bédirian, Simon Charbonneau, Victor Whitehead, Isabelle Collin, Jeffrey L. Cummings, and Howard Chertkow. 2005. “The Montreal Cognitive Assessment, MoCA: A Brief Screening Tool for Mild Cognitive Impairment.” *Journal of the American Geriatrics Society* 53 (4): 695–99. https://doi.org/10.1111/j.1532-5415.2005.53221.x.

Osterrieth, P. A. 1944. “Le Test de Copie d’une Figure Complexe.” *Arch Psychol* 30: 206–356.

Pergher, Valentina, Mahsa Alizadeh Shalchy, Anja Pahor, Marc M Van Hulle, Susanne M Jaeggi, and Aaron R Seitz. 2020. “Divergent Research Methods Limit Understanding of Working Memory Training.” *Journal of Cognitive Enhancement* 4 (1): 100–120. https://doi.org/10.1007/s41465-019-00134-7.

R Core Team. 2013. “R: A Language and Environment for Statistical Computing.” *R Foundation for Statistical Computing* Vienna (Austria).

Raven, J. C., and JH Court. 1996. *Standard Progressive Matrices: Sets A, B, C, D & E.* Oxford, England: Oxford Psychologists Press.

Redick, Thomas S. 2019. “The Hype Cycle of Working Memory Training.” *Current Directions in Psychological Science* 28 (5): 423–29. https://doi.org/10.1177/0963721419848668.

Redick, Thomas S., and Dakota R.B. Lindsey. 2013. “Complex Span and N-Back Measures of Working Memory: A Meta-Analysis.” *Psychonomic Bulletin and Review* 20 (6): 1102–13. https://doi.org/10.3758/s13423-013-0453-9.

Reitan, Ralph M. 1958. “Validity of the Trail Making Test as an Indicator of Organic Brain Damage.” *Perceptual and Motor Skills* 8 (3): 271–76. https://doi.org/10.2466/pms.1958.8.3.271.

Rey, André. 1941. *L’examen Psychologique Dans Les Cas d’encéphalopathie Traumatique.(Les Problems.)*. Archives de psychologie.

Rumpf, Ulrike, Inga Menze, Notger G. Müller, and Marlen Schmicker. 2019. “Investigating the Potential Role of Ecological Validity on Change-Detection Memory Tasks and Distractor Processing in Younger and Older Adults.” *Frontiers in Psychology* 10: 1046.

https://doi.org/10.3389/fpsyg.2019.01046.

Sala, Giovanni, N. Deniz Aksayli, K. Semir Tatlidil, Yasuyuki Gondo, and Fernand Gobet. 2019. “Working Memory Training Does Not Enhance Older Adults’ Cognitive Skills: A Comprehensive Meta-Analysis.” *Intelligence* 77: 101386. https://doi.org/10.1016/j.intell.2019.101386.

Sala, Sergio Della, Colin Gray, Alan Baddeley, Nadia Allamano, and Lindsey Wilson. 1999. “Pattern Span: A Tool for Unwelding Visuo-Spatial Memory.” *Neuropsychologia* 37 (10): 1189–99. https://doi.org/10.1016/S0028-3932(98)00159-6.

Sánchez-Cubillo, I., J. A. Periáñez, D. Adrover-Roig, J. M. Rodríguez-Sánchez, M. Ríos-Lago, J Tirapu, and F Barceló. 2009. “Construct Validity of the Trail Making Test: Role of Task-Switching, Working Memory, Inhibition/Interference Control, and Visuomotor Abilities.” *Journal of the International Neuropsychological Society*.

Scharfen, Jana, Katrin Jansen, and Heinz Holling. 2018. “Retest Effects in Working Memory Capacity Tests: A Meta-Analysis.” *Psychonomic Bulletin and Review* 25 (6): 2175–99. https://doi.org/10.3758/s13423-018-1461-6.

Schmicker, Marlen, Melanie Schwefel, Anne Katrin Vellage, and Notger G. Müller. 2016. “Training of Attentional Filtering, but Not of Memory Storage, Enhances Working Memory Efficiency by Strengthening the Neuronal Gatekeeper Network.” *Journal of Cognitive Neuroscience* 28 (4): 636–42. https://doi.org/10.1162/jocn_a_00922.

Schmidt, M. 1996. *Rey Aufitory Verbal Learning Test: A Handbook*. Los Angeles, CA: Westerm Psychological Services.

Shawn Green, C., Daphne Bavelier, Arthur F Kramer, Sophia Vinogradov, Ulrich Ansorge, Karlene K Ball, Ulrike Bingel, et al. 2019. “Improving Methodological Standards in Behavioral Interventions for Cognitive Enhancement.” *Journal of Cognitive Enhancement* 3 (1): 2–29. https://doi.org/10.1007/s41465-018-0115-y.

Shin, Eunsam, Hunjae Lee, Sang Ah Yoo, and Sang Chul Chong. 2015. “Training Improves the Capacity of Visual Working Memory When It Is Adaptive, Individualized, and Targeted.” Edited by Suliann Ben Hamed. *PLoS ONE* 10 (4): e0121702. https://doi.org/10.1371/journal.pone.0121702.

Soveri, Anna, Jan Antfolk, Linda Karlsson, Benny Salo, and Matti Laine. 2017. “Working Memory Training Revisited: A Multi-Level Meta-Analysis of n-Back Training Studies.” *Psychonomic Bulletin and Review* 24 (4): 1077–96.

Teixeira-Santos, Ana C., Célia S. Moreira, Rosana Magalhães, Carina Magalhães, Diana R.

Pereira, Jorge Leite, Sandra Carvalho, and Adriana Sampaio. 2019. “Reviewing Working Memory Training Gains in Healthy Older Adults: A Meta-Analytic Review of Transfer for Cognitive Outcomes.” *Neuroscience and Biobehavioral Reviews*. Elsevier Ltd. https://doi.org/10.1016/j.neubiorev.2019.05.009.

Wechsler, D. 2008. *Wechsler Adult Intelligence Scale–Fourth Edition (WAIS–IV)*. San Antonio, TX: The Psychological Corporation.

Weicker, Juliane, Nicole Hudl, Stefan Frisch, Jöran Lepsien, Karsten Mueller, Arno Villringer, and Angelika Thöne-Otto. 2018. “WOME: Theory-Based Working Memory Training - A Placebo-Controlled, Double-Blind Evaluation in Older Adults.” *Frontiers in Aging Neuroscience* 10. https://doi.org/10.3389/fnagi.2018.00247.

Weicker, Juliane, Arno Villringer, and Angelika Thöne-Otto. 2016. “Can Impaired Working Memory Functioning Be Improved by Training? A Meta-Analysis with a Special Focus on Brain Injured Patients.” *Neuropsychology* 30 (2): 190–212. https://doi.org/10.1037/neu0000227.

Table 1. Representation of training tasks according to each WM component of the multicomponent model (A. Baddeley 2012) and levels of difficulty for the MB and MB$^+$ WM training.

	Phonological loop (digit span)	Visuospatial Sketchpad (pattern span & corsi)		Central Executive (dual task)	Episodic Buffer (binding task)
MB	Increasing sequence length Adaptive	Increasing matrx size Adaptive	Increasing sequence length Adaptive	Increasing number of items Adaptive	Increasing number of probes Adaptive
MB+	MB + irrelevant noise effect + Similarity effect	MB + irrelevant picture effect	MB + irrelevant movement task	MB	MB

Notes. MB; model-based WM training, MB$^+$; model-based WM training with distractor inhibition.

Table 2. Demographic characteristics of the included sample at the pre-baseline.

	N	Sex (f, m)	Education (M±SD)	Age (M±SD)	MoCA (M±SD)	MADRS (M±SD)	Ishihara (M±SD)
MB$^+$ Training	29	17, 12	15.7 ± 3.5	64.3 ± 8.4	28.1 ± 1.3	0.7 ± 1.3	20.4 ± 3.8
MB Training	32	21 11	16.4 ± 4.0	64.5 ± 8.8	28.4 ± 1.1	0.6 ± 1.0	21.7 ± 0.7
Control Training	29	21, 8	15.3 ± 3.4	63.5 ± 8.1	28.3 ± 2.0	0.7 ± 1.1	21.9 ± 1.8
Dual n-back Training	33	19, 14	15.1 ± 3.2	64.0 ± 8.2	28.2 ± 1.2	0.9 ± 1.6	21.5 ± 1.1
Total	123	78, 45	15.6 ± 3.5	64.1 ± 8.2	28.3 ± 1.4	0.7 ± 1.2	21.4 ± 2.2

Table 3. Means (M), standard deviation (SD) and test statistics comparing pre-baseline and baseline session on all outcome measures on the whole group level.

	Pre-Baseline (M ± SD)	Baseline (M ± SD)	Pre-Baseline vs. Baseline (*p*)	Effect Size (*r*)
Complex Span Composite Score	50.65 ± 18.45	54.77 ± 19.38	< .001	0.37
Near Transfer				
ROCFT 30 min recall	20.83 ± 6.04	22.81 ± 5.56	< .001	0.39
RAVLT 30 min recall	10.98 ± 3.13	11.17 ± 2.84	n.s.	-
TMT B (s)	84.45 ± 30.22	75.69 ± 27.75	< .001	0.38
Far Transfer				
SPM	6.13 ± 2.13	6.06 ± 1.86	n.s.	-
DASS Depression	2.22 ± 3.28	1.64 ± 2.84	<.05	0.22
DASS Anxiety	1.44 ± 1.90	1.06 ± 1.76	<.05	0.21
DASS Stress	5.45 ± 4.69	4.67 ± 5.29	<.05	0.20
Other outcomes of Interest				
Digit Span	14.82 ± 3.46	15.07 ± 3.25	n.s.	-
Corsi Block	14.24 ± 3.36	14.59 ± 3.38	n.s.	-
n-back (wrong answers)	18.37 ± 9.02	15.88 ± 10.77	< .01	0.30

Note. ROCFT, the Rey Osterrieth Complex Figure Test; RAVLT, Rey Auditory Verbal Learning Test; TMT B, Trail Making Test form B; SPM, Standard Progressive Matrices; DASS, Depression Anxiety and Stress Scale.

Table 4. Means, standard deviations and univariate comparisons of the neuropsychological assessment at each session for each group.

	Pre-training	Post-training	Follow Up	Pre-vs Post-Training		Post-training vs Follow Up	
MB⁺				p	d	p	d
Complex Span	50.93 ± 17.74	60.59 ± 18.63	58.5 ± 20.16	.003*	.53	.815	.004
ROCFT 30 min	21.77 ± 6.41	25.09 ± 5.76	25.82 ± 4.66	.001*	.54	.717	.10
RAVLT 30 min	10.23 ± 2.62	10.41 ± 3.5	11.86 ± 1.99	.707	.06	.404	.18
TMT B (s)	78.95 ± 21.41	75.18 ± 14.82	71.33 ± 35.19	.335	.19	.892	.03
SPM	6.05 ± 2.01	6.5 ± 2.48	6.29 ± 1.9	.404	.19	.403	.29
DASS Depression	1.5 ± 3.28	1.82 ± 3.02	0.71 ± 1.44	.405	.10	.127	.36
DASS Anxiety	0.5 ± 0.8	0.91 ± 2.04	0.64 ± 1.15	.323	.24	.755	.11
DASS Stress	3.41 ± 3.29	4.27 ± 3.99	2.07 ± 2.2	.325	.23	.069	.62
n-back	17.68 ± 12.56	13.41 ± 10.34	13.86 ± 12.01	.102	.37	.542	.08
Digit Span	14.14 ± 2.59	14.27 ± 3.37	15.07 ± 3.41	.792	.04	.865	.04
Corsi Block	14.45 ± 3.65	14.64 ± 3.47	15.21 ± 3.79	.786	.05	.303	.19
MB							
Complex Span	58.83 ± 20.52	59.58 ± 16.65	60 ± 18.64	.796	.04	.459	.18
ROCFT 30 min	21.73 ± 5.76	23.23 ± 6.37	23.47 ± 5.35	.278	.25	.904	.03
RAVLT 30 min	10.96 ± 3.33	11.54 ± 3.4	11.93 ± 2.52	.262	.17	.224	.22
TMT B (s)	76.91 ± 33.74	65.08 ± 15.83	61.31 ± 18.48	.039*	.38	.389	.17
SPM	6.12 ± 1.48	5.88 ± 1.83	6.07 ± 1.39	.552	.15	.922	.04
DASS Depression	1.21 ± 1.61	0.82 ± 1.3	1.27 ± 1.71	.162	.26	.351	.27
DASS Anxiety	1.71 ± 2.44	1.59 ± 2.5	2.13 ± 3.46	.783	.05	.701	.04
DASS Stress	4.42 ± 5.52	4.51 ± 6.02	3.93 ± 5.69	.889	.02	.930	.01
n-back	19.21 ± 11.52	15.38 ± 8.47	14.73 ± 9.82	.045*	.36	.797	.04
Digit Span	16.08 ± 3.19	16.42 ± 3.61	17.13 ± 2.92	.569	.10	.655	.09
Corsi Block	14.83 ± 3.47	14.79 ± 3.45	15.4 ± 2.64	.943	.01	.8	.06
Dual n-back							
Complex Span	53.16 ± 19.93	55.81 ± 18.54	53.25 ± 15.74	.237	.14	.581	.07
ROCFT 30 min	23.25 ± 5.73	24.17 ± 5.18	26.41 ± 3.53	.386	.17	.074	.60
RAVLT 30 min	11.54 ± 3.02	12.04 ± 2.51	12.19 ± 2.4	.306	.18	.928	.02
TMT B (s)	69.49 ± 21.76	80.4 ± 37.41	68.31 ± 16.99	.099	.34	.420	.23
SPM	6.19 ± 1.98	6.12 ± 2.05	6.62 ± 1.67	.859	.04	.060	.52
DASS Depression	1.58 ± 2.53	1.73 ± 2.81	1.62 ± 2.8	.733	.06	.508	.14
DASS Anxiety	0.65 ± 1.23	0.92 ± 1.16	1.19 ± 2.32	.215	.22	.383	.17
DASS Stress	5.58 ± 6.33	4.77 ± 4.87	5.38 ± 5.23	.337	.14	.868	.02
n-back	14.81 ± 9.83	7.92 ± 8.86	9 ± 11.05	.001*	.73	.570	.19
Digit Span	15.23 ± 3.46	15.54 ± 3.4	15.56 ± 2.78	.448	.09	.734	.06
Corsi Block	14.38 ± 3.51	15.81 ± 3.26	15.88 ± 3.38	.070	.42	.417	.29
Control							
Complex Span	56.68 ± 18.69	59.61 ± 16.08	55.47 ± 12.18	.357	.17	.022*	.32
ROCFT 30 min	23 ± 4.18	23.13 ± 7.59	21.73 ± 6.36	.902	.02	.254	.20
RAVLT 30 min	11.47 ± 2.2	12.53 ± 2.29	11.93 ± 2.49	.099	.47	.818	.06
TMT B (s)	77.99 ± 28.22	82.21 ± 44.81	71.48 ± 30.31	.603	.10	.056	.28
SPM	6.26 ± 2.02	6.37 ± 1.92	6.27 ± 2.05	.816	.05	1	.00
DASS Depression	1.47 ± 2.48	2.21 ± 4.04	2.93 ± 6.25	.163	.17	.048*	.16
DASS Anxiety	1.26 ± 2.08	1.16 ± 2.65	1.87 ± 2.97	.695	.04	.017*	.54
DASS Stress	4.11 ± 3.4	4.16 ± 5.21	5.27 ± 5.7	.946	.01	.127	.16
n-back	12.16 ± 7.89	11.42 ± 8.27	16.13 ± 12.37	.618	.09	.242	.31
Digit Span	14.68 ± 2.93	15.42 ± 3.1	15.6 ± 3.62	.206	.24	.768	.06
Corsi Block	14.47 ± 3.52	14.68 ± 3.97	14.07 ± 3.49	.805	.06	.183	.17

Note. °, significance level at $p < .1$, *, significance level at $p < .05$

Table 5. Group x Session interaction effects of the linear mixed model for all WM, near and far transfer measures with MB$^+$ training as a priori contrast.

	Delta Pre/Post-Training		**Delta Post-Training/FU**	
	χ^2	p	χ^2	p
Complex Span Composite Score	7.29	.06°	2.99	.39
Near Transfer				
ROCFT 30 min recall	4.38	.22	5.96	.11
RAVLT 30 min recall	1.84	.61	1.89	.59
TMT B (s)	8.06	.04*	2.91	.41
Far Transfer				
SPM	1.18	.76	2.59	.46
DASS Depression	4.93	.18	5.35	.15
DASS Anxiety	1.57	.66	3.59	.31
DASS Stress	2.09	.55	8.04	.04*
Other outcomes of Interest				
Digit Span	0.97	.81	0.49	.92
Corsi Block	2.46	.48	1.21	.75
n-back (wrong answers)	4.15	.25	1.66	.64

Note. °, significance level at $p < .1$, *, significance level at $p < .05$

Figure Captions

Fig 1. Overview of study design.

Fig 2. Flowchart of participant recruitment and study inclusion.

Fig 3. Mean and standard error of the complex span composite score for each the model-based (MB), control, dual-n-back and model-based plus (MB$^+$) training group over the four sessions.

Fig 4. Representation of mean difference of the complex span composite score between pre- and post-training in all training groups.

Supplementary Material

9 Supplementary Data

Construction of the MB and MB^+ training

Phonological Loop Task. Previous literature suggests that immediate serial recall tests using a small set of digits, letters or unrelated short words are used for the assessment of the phonological loop (67). In these simple span tests, participants hear a short sequence of digits, letters or words which they have to repeat in the correct order. For our phonological training task, thus a simple letter span test was used as the basic paradigm. Since it has been shown that the immediate memory span declines as the item length increases, the number of presented digits is used as a level of difficulty in the MB training. We recorded all letters of the alphabet in German language. Based on the pronunciation of the letters in German, we included the letters A, B, C, D, E, F, G, I, L, M, O, P, R, S, T, U, W, Z in the general letter pool. A random sequence of letters was presented phonologically over headphones and participants were instructed to recall this sequence. After a retention phase of 2.5 s, participants were shown tabs with the written letters in order to enter the heard sequence. There was no time limit for entering the sequence, however, as soon as the correct number of letters was entered, the keyboard disappeared and after a 1 s break a next sequence was phonologically presented. For the MB training, the sequence length started at 1 letter and increased with one additional letter after 3 repetitions of the original sequence length.

However, it has been extensively documented that similar items (E, G, B, D) are less accurately recalled than dissimilar items (A, F, G, U) also known as the "similarity effect" and that irrelevant sounds disturb the recall of the items, known as the "irrelevant noise effect" (35). Although it has been shown that a training on dichotic listening reduced task-irrelevant speech interference (68), no training up to date included task levels based on the irrelevant noise or similarity effect. Thus, the phonological loop training task for the MB^+ training consisted next to the increased sequence length of the following two difficulty levels: 1) presence of irrelevant background sounds or not and 2) presentation of dissimilar or similar digits of items. The irrelevant background noises consisted of royalty free "space" sounds (https:/freesound.org) which were selected with the aim of increasing interference to the letter span task. For the similarity level, a second pool of letters was created which included letters that are perceived as similar in German (B, C, D, E, G, P, T, W or K, H, A). An easy level would therefore start by a small sequence length and dissimilar digits presented without irrelevant sounds. Subsequently, after the presentation of two sequences with dissimilar letters, one sequence was presented with an irrelevant noise during the presentation of the sequence and the retention phase. As a next level, participants heard a sequence of similar letters and additionally the irrelevant noise. The levels of similar letters or irrelevant noises or both are added to the increasing number of letters to induce increasing difficulty of the training.

Visuospatial Sketchpad Tasks. It has been suggested, that the visuospatial sketchpad can further be distinguished in a visual and spatial subcomponent. For this reason, two tasks were implemented to train the visuospatial sketchpad component. Our review showed that visual pattern span test is broadly used to assess the visual subcomponent (36) and was thus seen as the basis for our MB and MB$^+$ training task. In the pattern span task, a grid with colored and uncolored squares is presented to the participants for a short time span in order to memorize the pattern of colored squares. Participants then have to recall the pattern by filling all squares that were colored in the previous grid in the right positions. This was done accordingly in our training, by presenting a 2x2 (minimum) and with increasing difficulty up to 5x6 square grid (maximum) for 3 seconds. After a short retention phase of 2.5 seconds, an empty grid appeared to fill in the memorized colored squares. Regarding the spatial subcomponent of the visuospatial sketchpad, the most common task used for assessment is the Corsi block tapping test (35,37), which therefore built the basis for our training task of the spatial subcomponent. Visuospatial WM is tested by a tapping-sequence of pegs arranged on a board. Subsequently, participants have to recall the sequence by tapping on the right pegs. In our MB and MB$^+$ trainings, objects were arranged in a square and flashed in a specific sequence which the participant had to recall in the same order by tapping on the objects. The task improves in difficulty by adding an item to the previous sequence which challenges the memory recall of the sequence.

Already in early experiments it has been shown, that the presentation of an irrelevant picture after the pattern span matrix and during the retention phase leads to a reduction in performance on the pattern recall, suggesting that the passive visual store is accessible by presented interfering visual inputs (69). It has also been shown, that training of filtering efficiency can improve visual WM, however a WM training task combining filtering efficiency and WM does not exist to date (58). Hence, next to the increasing size of the matrix and number of filled squares as done in the MB training, a level of difficulty was created for the MB$^+$ by adding an irrelevant visually loaded picture during the retention phase and the recall of the grid squares. Six pictures were selected from the NASA image and video library based on their visual features since it has been described that the content of the picture (densitiy, color palette, unspecifity of content?) influences the irrelevant picture effect. Similar to the pattern span task, it has been shown that an irrelevant task of haptic movement shifts the spatial attention and by that interferes with a spatial WM task such as the Corsi block tapping test (70,71). As an additional level of difficulty for the MB$^+$ training, we therefore added a haptic irrelevant movement task between the presentation and recall of the Corsi task next to the increasing sequence length of the presented items. Following the presentation of the object sequence, participants were asked to follow lines displayed as star constellations with their finger until the lines disappeared. When the whole figure disappeared, the object arrangement appeared again and they were asked to recall the sequence of the highlighted objects.

Central Executive Task. The central executive has been postulated to be the most important, however least investigated component of the multicomponent model. It has been investigated that the central executive covers four basic capacities: The ability to focus, divide and switch attention as well as the ability to relate content of WM to long-term memory. Due to its importance in the theoretical framework, we developed a task to train the coordinative function of WM. Random word, letter or digit generation tasks or as well dual tasks have been suggested to assess and investigate the central executive (38,39). Random digit generation tasks showed, that by increasing speed of generation the randomness decreases and it therefore has been implied, that the non-randomness seems to include information on the limit of processing capacity. Due to the nature of this kind of task, it seems unlikely to build a training task with levels. Hence, we developed a basic dual task which targets two tasks at the same time, in our case visuospatial sketchpad and the phonological loop, and took this as the basis of the central executive training task. Subjects were presented a specific item which they had to hold in mind and search in a pool of distracting items. With the right hand, they were instructed to tab on the items they had to remember. With increasing difficulty, more distracting items were shown and the speed of presentation increased. At the same time, they were hearing a high or low tone. By placing the left hand on two buttons, they had to indicate which tone they heard while additionally the speed of presentation increased with increasing level.

Episodic Buffer Task. Since it has been shown that the relation of content of WM to long term memory is not only a process which involves the central executive and can rather be explained through binding procedures, the episodic buffer was introduced to the model. The episodic buffer is a separate storage system of limited capacity using a multimodal code by providing a temporary retention of integrated information (72). The episodic part holds information from other cognitive systems inclusive the WM in so called “scenes or episodes”. It is a buffer, as it displays an intermediary between subsystems with a different code. A review came to the conclusion, that the episodic buffer can be investigated using unimodal or cross modal binding tasks (73). Typically, tasks where letters or words and a spatial location are presented concurrently are used. In our MB and MB^{+} training, a spatial order of letters on certain position with or without a shape around them are presented on a stimulus slide. Followed by a blank slide, one letter or word on a random position is presented and participants had to answer, if 1) the word or letter is the same then one on the stimulus slide and 2) if the location is on the same position than on the stimulus slide. The participant has to give an answer only, if both questions can be answered as correct (72,74). This task was used as the basis for the episodic buffer training task. In our training we used letters framed by squares. In order to create levels, the stimulus array was increased by a letter with increasing difficulty.

5.2 Additive and Interaction Effects of Working Memory and Motor Sequence Trainings on Brain Functional Connectivity

Additive and interaction effects of working memory and motor sequence training on brain functional connectivity.

Priska Zuber[1], Laura Gaetano[2], Alessandra Griffa[3,4], Manuel Huerbin[5], Ludovico Pedullà[6,7], Laura Bonzano[7,8], Anna Altermatt[5], Charidimos Tsagkas[9,10], Katrin Parmar[9-11], Patric Hagmann[4,12], Jens Wuerfel[5,13], Ludwig Kappos[9,10,14], Till Sprenger[15], Olaf Sporns[16,17], Stefano Magon[2,9]

1. Division of Cognitive Neuroscience, Faculty of Psychology, University of Basel, Switzerland
2. Roche Pharma Research and Early Development, Roche Innovation Center Basel, F. Hoffmann-La Roche Ltd., Basel, Switzerland
3. Department of Clinical Neurosciences, Division of Neurology, Geneva University Hospitals and Faculty of Medicine, University of Geneva, Geneva, Switzerland
4. Institute of Bioengineering, Center of Neuroprosthetics, École Polytechnique Fédérale De Lausanne (EPFL), Geneva, Switzerland
5. Medical Image Analysis Center (MIAC AG), Basel, Switzerland
6. Department of Experimental Medicine, Section of Human Physiology, University of Genoa, Genoa, Italy
7. Italian Multiple Sclerosis Foundation, Scientific Research Area, Genoa, Italy,
8. Department of Neuroscience, Rehabilitation, Ophthalmology, Genetics, Maternal and Child Health, University of Genoa, Genoa, Italy
9. Neurologic Clinic and Policlinic, Departments of Medicine, Clinical Research and Biomedical Engineering, University Hospital Basel and University of Basel, Basel, Switzerland
10. Translational Imaging in Neurology (ThINk) Basel, Department of Medicine and Biomedical Engineering, University Hospital Basel and University of Basel, Basel, Switzerland
11. Reha Rheinfelden, Rheinfelden, Switzerland
12. Department of Radiology, Centre Hospitalier Universitaire Vaudois (CHUV) and University of Lausanne (UNIL), Switzerland
13. Department of Biomedical Engineering, University of Basel, Switzerland
14. Research Center for Clinical Neuroimmunology and Neuroscience Basel (RC2NB), Departments of Medicine, Clinical Research and Biomedical Engineering, University Hospital Basel and University of Basel, Basel, Switzerland
15. Department of Neurology, DKD Helios Klinik Wiesbaden, Germany
16. Department of Psychological and Brain Sciences, Indiana University, Bloomington, Indiana, USA
17. Indiana University Network Science Institute, Indiana University, Bloomington, Indiana, USA

Corresponding Author

Stefano Magon PhD

Email: stefano.magon@roche.com

Postal address: Department of Neurology, University Hospital Basel, Petersgraben 4, 4031 Basel

Declaration of Interest: None. Hoffman-La Roche Ltd. was not involved in the research.

Highlights

- Working memory and motor training increased resting state functional connectivity
- Connectivity was mainly increased in right parietal and left temporal lobes
- The strength of connectivity was modulated by the order of training administration

Abstract

Background: Although it has been suggested that working memory (WM) and motor sequence learning (MSL) share behavioral and neural mechanisms, the additive and interactive effects of training have not been studied.
Objectives: To study chantes in brain functional connectivity (FC) induced by two sequential training programs (WM+MSL and MSL+WM) and a combined one (WMxMSL).
Methods: 54 healthy subjects (27 women; mean age: 30.2±8.6 years) were allocated to three training groups and underwent twenty-four 40-min training sessions over six weeks and four cognitive testing sessions including functional MRI. The first two sessions served as double-baseline to account for practice effects. Test performances were compared using linear mixed-effects models and t tests. Resting state fMRI data were analysed using FSL.
Results: Processing speed, verbal WM and manual dexterity increased following training in all groups. MSL+WM training led to additive effects in processing speed and verbal WM. Increased FC was found after training in a complex network involving the right angular gyrus, bank of the left superior temporal sulcus, right superior parietal gyrus, bilateral middle temporal gyri and left precentral gyrus. No difference in FC was found between the two individual baseline sessions.
Conclusion: This study indicates distinct patterns of resting state FC modulation related to sequential and combined WM and MSL training suggesting a relevance of the order in which trainings are performed. These observations could provide new insight for the planning of effective training/rehabilitation programs.

Keywords

Working Memory, Motor Sequence Learning, Training, Functional Connectivity, fMRI

1. Background

Motor learning describes the human ability to acquire new motor skills (1) and has been defined as "[…] increased spatial and temporal accuracy of movement with practice." (2). Learning new motor skills on the one hand results from explicit learning, where the learner relies on his declarative memory by applying performance rules in order to acquire and control a new set of movements. On the other hand, acquiring new skills can also occur implicitly through repetition of movements with practice based on visual or tactile sensory feedback, released from explicit memory control (3). Supporting a distinction in explicit and implicit learning of movements, it has been demonstrated that, after damage of the medial temporal lobe due to brain injury or stroke, the capacity of implicit motor sequence learning (MSL) can be retained, whereas the capacity of explicit learning of motor skills is impaired (4). Therefore, MSL could be an interesting function to target in the context of training and rehabilitation, since it has been hypothesized that MSL is not a purely motor function and that cognitive functions such as working memory are involved already in early stages of MSL (2,5).

Working memory (WM) refers to a cognitive system which temporarily maintains, stores and manipulates information and acts as an interface between perception, long-term memory and action (6,7). Following Baddeley's multicomponent model, WM can be divided into a visuospatial, a verbal, a coordinative and a buffer component (7). Since WM is centrally involved in numerous higher order cognitive functions (8) and plays a role in age-related cognitive decline (9), WM training has been suggested to improve WM capacity, suggesting that it may ameliorate cognitive decline (10). WM trainings gained huge interest over the last decade and numerous studies showed their efficacy in improving WM capacity as well as transfer effects to cognitive domains that are not specifically targeted by the training (11,12).

There is evidence that MSL is related to WM abilities. It has been shown that the spatial component of WM and visuomotor adaptation share common processes (5) and the individual spatial WM performance predicts the rate of implicit MSL (13). Furthermore, a correlation between visual and visuospatial WM with reaction time change in a serial reaction time task has been described which further supports a sharing component between WM and implicit MSL (14). These shared components are also mirrored in the neural activity. Functional magnetic resonance imaging (fMRI) evidence indicates that brain regions conventionally associated with motor activation show sub-regions that contribute to motor and cognitive processes jointly (15). To that end, the right dorsolateral prefrontal cortex, parietal and premotor regions, basal ganglia as well as cerebellar areas have been described to be involved in MSL as well as WM performance (13,15). Regarding brain functional connectivity (FC), dynamic changes in terms of integration within and between the premotor and sensorimotor network have been shown to be associated with MSL. Moreover, it has been shown that this functional integration decreases with practice over a four-week training of motor learning (16). Regarding WM and motor training studies, a meta-analysis described that across 53 studies showing brain activation decreases in numerous brain regions solely the dorsolateral prefrontal cortex showed a consistent activation decrease. Additionally, training-related increases were consistent in the salience-network (supplementary motor area, anterior cingulate cortex, and inferior frontal gyrus, anterior insula), dorsal attention network (the superior parietal cortex, intraparietal sulcus, frontal eyefield), striatum, thalamus, ventral and dorsal visual and superior temporal cortices (17). Whereas these regions showed FC changes during task-fMRI, recent studies further indicate changes in resting state FC following WM training (18) but also after motor training (19).

Despite the reported relationship between WM and MSL, to our knowledge there are currently no studies that investigate the interactive effects of MSL and WM training on brain activation patterns or behavioral outcomes. It is, therefore, necessary to investigate the sequential order of training administration, in order to study the interactive neural and behavioral effects of MSL and WM training. Thus, the present study aimed to investigate how brain FC and behavioural outcomes are modulated by sequential training programs with varying order of administration (WM + MSL and MSL + WM) or combined administration (WM and MSL in the same session). In the first setting, the purpose was to quantify the additive effects, while, in the second case, the interactive ones.

2. Methods

2.1 Participants

Fifty-four healthy subjects were included in the study (27 women and 27 men; mean age: 30.8±8.5, age range: 20-51 years). Inclusion criteria were: age between 18 and 65 years old, right-handedness (above the 5^{th} decile) according to the extended version of the Edinburgh handedness questionnaire (20) as well as no history of neurological, psychiatric disorders or substances abuse. Written informed consent was obtained from each participant after a detailed explanation of the study procedures. The study was approved by the local ethics committee (Ethikkommission Nordwest und Zentralschweiz) and was conducted in accordance with the Declaration of Helsinki.

2.2 Experimental design

All participants were randomly assigned to three groups (A, B and C) using the minimization approach in order to create comparable groups in terms of age and gender (see table 1). All groups underwent a WM training (WMT) and a MSL training (MSLT), both consisting of 12 sessions (four times a week). The WMT's session lasted for 25 minutes and the MSLT's

sessions lasted for 15 minutes. The group A and group B performed the two trainings sequentially, whereas the second training started after the completion of the 12 sessions of the first one. The two trainings were presented with a different order for the two groups: WMT+MSLT for the group A and MSLT+WMT for the group B. This design allowed to investigate the additive effects of the two trainings. The group C performed both trainings during the same training session (for 12 sessions, each with a total duration of 40 minutes) in order to investigate the interaction effects (WMTxMSLT).
Additionally, participants underwent a first baseline magnetic resonance imaging (MRI) three weeks before the first training session (BL1) and a second baseline MRI within two days before the first training session (BL2, figure 1). The dual baseline approach has been employed to assess the repetition effect of multiple MRIs on the FC changes as well as neuropsychological test performance. All participants underwent a third MRI within one week from the end of the first training program (T3). Groups A and B underwent a fourth MRI after the second training period (T4, within one week; figure 1).

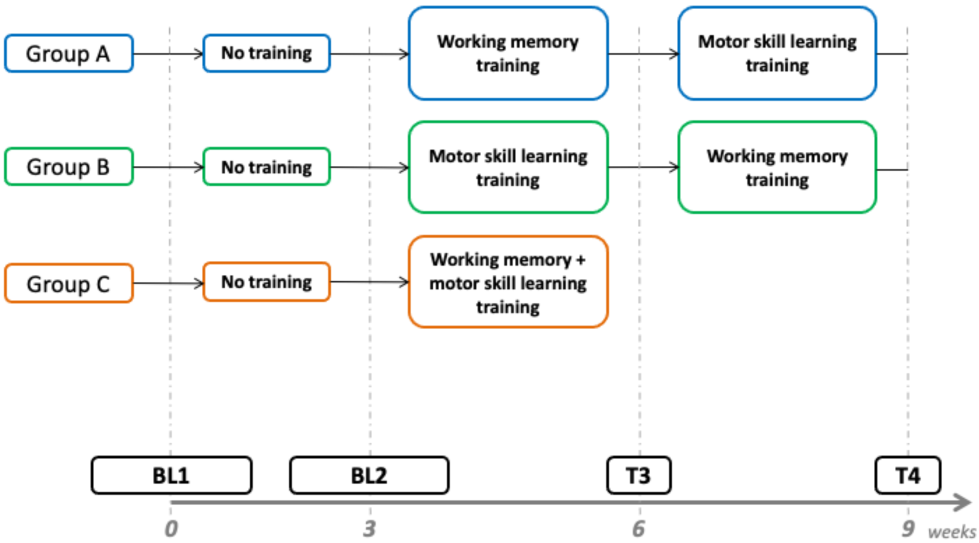

Figure 1. Study procedures. Each assessment (BL 1, BL2, T3 and T4) included an MRI and cognitive testing.

2.3 Training

All training sessions have been performed at home using touch-screen tablet devices provided by the research group. The trainings were developed using Java in Android Studio v1.5.1 (https://developer.android.com/studio/index.html) and downloaded on Lenovo TAB A10 with Android 4.4 as operating system. Both trainings were based on an adaptive design. Therefore, the difficulties of the tasks were modulated according the participant’s performance.

The training for MSL was developed in-house based on a single-paradigm design. The participants learned the procedural knowledge needed to execute a series of actions following a cue presented on the screen. Specifically, four circles were presented on the screen, each one associated with a finger of the dominant hand, from left (index finger) to right (pinkie). The thumb was not included in the sequence. The participants were asked to touch the circle that changed colour with the corresponding finger (figure 2). The levels were based on the number of items included in the sequence (the first level included 5 items) and on the interstimulus interval (ISI), i.e. 1200 ms, 900 ms and 600 ms. For every level, the sequence of items was repeated 10 times. The level of difficulty was increased stepwise, if a) the accuracy exceeded 80% and b) reaction time of the correct answers on the last five repetitions was faster compared to the first five repetitions on average. If the participant failed to increase the level of difficulty for three consecutive repetitions of the same level, the difficulty was decreased by one step.

The COGNI-TRAcK (21–23) was used to train the WM skills (figure 3). Briefly, it includes the following three trainings. 1) The visuospatial training (WMT-VS), in which circles were presented one at a time in a three-by-three grid-like interface. Participants had to remember the location and the order of the stimuli. Levels were defined by the number of stimuli (the first level included four elements) and by the ISI, i.e. 2000 ms, 1500 ms and 1000 ms. The difficulty was increased if the accuracy was 100% for the levels with less than eight stimuli and 80% for higher number of stimuli. 2) Operation N-back training (WMT-OP), in which paired of numbers (from 1 to 4) were showed in a random sequence on the screen (e.g., 3+4). Participants were asked to memorize the sum of the two numbers presented (ranging from 2 to 8) and select the correct answer on the screen, which referred to N stimuli ago. The levels were defined by the value of N and by the ISI (from 5000 ms to 3000 ms in steps of 500 ms). The difficulty was changed if the accuracy was higher than 80%. 3) Dual N-back training (WMT-DT), in which the stimuli (numbers for 1 to 4) were randomly presented one at a time in one of the four possible positions along a line. The participants were asked to memorize the number and the position. Then, they were also asked to select the correct item on screen using the left hand (1, 2, 3 or 4), and the correct position of the stimulus appeared N stimuli ago using the right hand. The definition of the level remained the same as for the previous task. The change of the level occurred if the accuracy was higher than 75%.

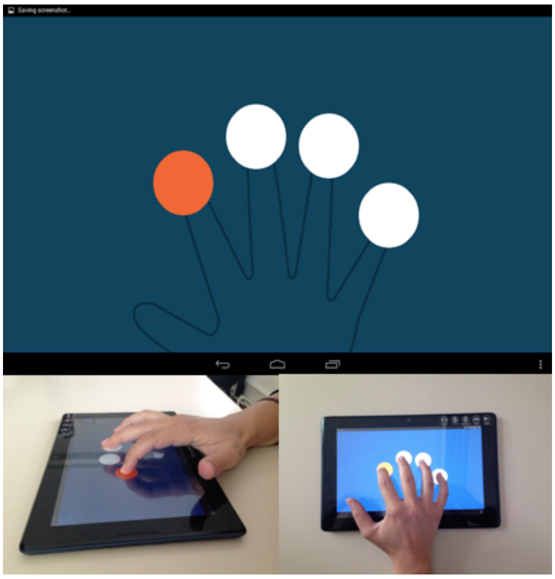

Figure 2. MSL training on the tablet device.

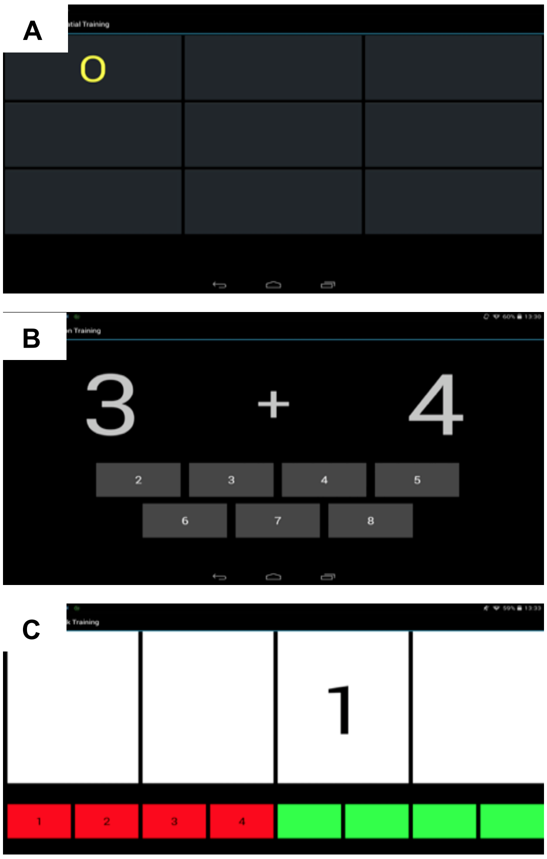

Figure 3. CogniTrack WM training with A) the visuospatial, B) the operation n-back task and C) the dual-n-back task.

2.4 Cognitive Assessment

At all four MRIs (BL1, BL2, T3 and T4), an additional cognitive assessment was performed. All participants underwent neuropsychological assessment addressing attention, processing speed and working memory using both paper/pencil and computerized tests. Namely, information processing speed was assessed using the Paced Auditory Serial Addition Task (PASAT (24)) and the oral version of the Symbol Digit Modalities Test (SDMT (25)), verbal WM was assessed using the forward and backward digit span (WAIS-IV (26)) and visuospatial WM was assessed with the Corsi Block Tapping Test (27) and a visuospatial n-back task. Additionally, finger dexterity was assessed using the nine-hole peg test (9HPT (28)), performed for the right and left hand. The 30-minute assessments were performed at the Department of Neurology, University Hospital Basel by trained psychologists.

2.5 MRI protocol

MRI data were collected on a 3 Tesla scanner (Siemens Prisma) using a 64-channel head coil. The MRI protocol included a 3D T1-weighted (T1w; MPRAGE, TR= 2400 ms, TE=2.32 ms, TI=1100 ms, voxel, flip angle=8°, number of slides=256, voxel size=0.7 mm^3 isotropic), and a multi-band accelerated echo-planar imaging (EPI) sequence (29) used to collect the FC data (TR = 768 ms, TE = 37 ms, flip angle = 52°, bandwidth = 2290 Hz/Px, multi-band accelerator factor = 8, number of slides = 72, voxel size = 2 mm^3 isotropic, phase-encoding = anterior-posterior, number of acquisition = 1160). Six additional images, three with anterior–posterior and three with posterior-anterior phase-encoding direction were collected with the same parameters in order to correct for EPI distortions (30). MRI data of all participants was checked for incidental findings by a board certified radiologist.

2.6 Brain parcellation

Parcellation of cortical regions and subcortical nuclei was performed using Freesurfer (Version 5.3; (31)). The cortical parcellation was based on the Desikan-Killiany atlas (32) and the subcortical parcellation was done using the atlas implemented in Freesurfer (33). Higher resolution cortical parcellation (234 cortical regions) was generated from the Desikan-Killiany parcellation using the connectome mapper toolbox that included the Lausanne 2008 atlas (34). Additionally, the cerebellum was segmented in 15 subregions for each hemisphere using the rapid automatic segmentation of the human cerebellum and its lobules (RASCAL) algorithm (35). The final parcellation used to generate the connectivity matrices included 264 brain regions (212 cortical, 22 subcortical and 30 cerebellar ROIs) (36).

2.7 Functional connectivity analysis

The fMRI volumes were preprocessed using FSL (37) with the following steps: 1) motion correction; 2) distortion correction using FSL-topup (38); 3) temporal filtering to remove physiological noise (band-pass filter = 0.01 – 0.08 Hz); 4) regression of the white matter signal, cerebrospinal fluid signal and estimated motion parameters including the outliers identified by the FSL motion outliears tool.

For every MRI session and subject, the segmented T1w images were then registered to the lower resolution EPI images. For every segmented brain region, the signal of the preprocessed EPI was averaged. The FC matrices were generated by computing the cross-correlations among the averaged signals of all 264 regions. The brain regions were rearranged according to a set of canonical resting state networks as described by (39).

2.8 Statistical analysis

Demographic factors were compared among groups using the t-test and chi-square test. Baseline differences were analyzed using an ANOVA model with performance at BL1 or BL2 as outcome and group as the between group factor. Repetition effects between BL1 and BL2 were analyzed using linear models on the whole group level with session as within-subjects factor on neuropsychological test performance. Neuropsychological data were analyzed using a linear mixed model with test performance as outcome, the interaction term session x training group as fixed effect and study participant as random effect. χ^2 statistics were gained by running an ANOVA over the linear model using type II sums of squares. All data were analyzed in R Studio, Version 1.2.1335 (40).

Differences in the strength of connectivity between sessions were assessed in the whole sample using the network-based statistic algorithm (NBS, (41)) as implemented in the brain connectivity toolbox (42). The statistical threshould estimated using permutation testing (number of permutation: 10000) was set at 0.05 with a t-value of four. The BL1 MRI was compared to the BL2 MRI and to the T4 MRI .

Differences among subgroups were investigated by computing the effect size (Cohen's *d*) of the differences of the FC strength among MRI sessions for each pair of nodes belonging to the network identified in the whole sample analysis by the NBS algorithm.

3. Results

3.1 Behavioral Results

3.1.1 Baseline characteristics of training groups

No differences were observed among groups in terms of demographic characteristics, namely sex and age (table 1). When performing the trainings separately (Group A and B), participants did slightly more training sessions of the MSL than the WM training, independent of the order the training was given. The group that completed all trainings together, however, did approximately the same amount of sessions on both trainings (table 1). Mean, standard deviation and range of reached levels for each training task of the different groups are displayed in table 2.

Table 1. Demographic characteristics and training adherence for the three groups

	Age, years (M ± SD)	Sex (f/m)	MSL training sessions, n (M ± SD)	WM training sessions, n (M ± SD)
Group A	30.1 ± 8.8	9/9	12.2 ± 3.75	10.8 ± 3.0
Group B	30.6 ± 8.4	9/9	13.7 ± 3.29	12.0 ± 2.59
Group C	30.1 ± 8.9	9/9	11.7 ± 3.12	12.2 ± 1.96

Notes. M = Mean; SD = Standard Deviation; n: number.

Table 2. Mean (M), standard deviation (SD) and range of reached levels for WM and MSL (MOST) at the end of the training.

	WM-OP		WM-VS		WM-DT		MSL	
	M±SD	Range	M±SD	Range	M±SD	Range	M±SD	Range
Group A	22.4±5.5	9-30	17±4.7	10-27	17.3±3.3	10-22	29.4±7.2	11-38
Group B	23.6±3.8	15-30	19.2±5.6	12-36	18.7±2.9	15-23	32.6±7.7	9-42
Group C	23.9±5	11-32	19.4±6.9	10-41	18.1±3.1	10-23	30.6±7.31	15-38

Notes. OP = Operation n-back task; VS = Visuospatial task; DT = Dual n-back task.

3.1.2 Practice effects between BL1 and BL2

The analyses indicate no differences between groups in their performance on all tasks at both the BL1 and BL2. The whole-group analyses performed to investigate possible practice effects from BL1 to BL2 resulted in a significant session effect in SDMT ($\chi^2(1)$ = 61.63, p < .001), PASAT ($\chi^2(1)$ = 12.92, p < .001), digit span test ($\chi^2(1)$ = 5.21, p < .05), and 9HPT on the non-dominant left hand ($\chi^2(1)$ = 6.69 , p < .05), with, in all cases, an increase in performance at BL2 compared to BL1 (SDMT: b = 7.07, t(53) = 7.78, p < .001, r = 0.69; PASAT: b = 2.59, t(53) = 3.56, p < .001, r = 0.44; digit span test: b = 0.70, t(53) = 2.26, p < .05, r = 0.29; 9HPT of the non-dominant left hand: b = -0.8, t(53) = -2.56, p < .05, r = 0.33). Instead, there were no changes between BL1 and BL2 on the whole group level in the Corsi block tapping test ($\chi^2(1)$ = 2.56, p = .11) and in the 9HPT of the dominant right hand ($\chi^2(1)$ = 3.58, p = .06).

3.1.3 Training effects

The linear mixed effects models showed a significant SDMT performance improvement between sessions ($\chi^2(2)$ = 71.09, p < .001), both at T3 compared to BL2 and T4 compared to T3. A significant session effect was found in the PASAT ($\chi^2(2)$ = 28.26, p < .001) with an increase in performance at T3 compared to BL2 and tendency for significance between T4 compared to T3. Further, a significant digit span performance improvement between sessions ($\chi^2(2)$ = 12.78, p < .05) was found in both at T3 compared to BL2 and at T4 compared to T3. Finally, a significant session effect was found in the 9HPT right hand ($\chi^2(2)$ = 12.81, p < .05) with increased performance at both T3 compared to BL2 and T4 compared to T3 (table 3). For the left hand of the 9HPT, there were no significant session, group or session by group effects. The results revealed a tendency for significance in the group by session interaction in the SDMT ($\chi^2(4)$ = 9.01, p = .06). However, the *post-hoc* analyses yielded no significant results in the contrasts of the SDMT group by session interaction. For all other tests, neither group nor the group x session interaction was significant.

Table 3. Significant contrasts of linear models for session effect between BL2 vs T3 and T3 vs T4.

	BL2 vs. T3					**T3 vs. T4**				
	b	**df**	**t**	**p**	**r**	**b**	**df**	**t**	**p**	**r**
Session Effect										
SDMT	-5.76	102	-4.76	<.001	.43	-5.43	102	-5.95	<.001	.51
PASAT	-2.98	102	-4.82	<.001	.43	-2.11	102	-2.33	.06	.23
digit span test	-1.07	102	-3.37	<.05	32	-0.74	102	-2.57	<.05	.25
9HPT right hand	0.67	102	3.14	< .05	.29	0.59	102	2.92	<.05	.28

Note. SDMT; Symbol Digit Modalities Test, PASAT; Paced Auditory Serial Addition Test, 9HPT; Nine Hole Peg Test.

The univariate analyses within group A (WM+MSL) revealed a significant improvement in performance from BL2 to T3 in the SDMT (t(17) = -3.09, p < 0.05). All other comparisons for the other tests showed no significant changes. For the group B (MSL+WM), the univariate analyses showed significant improvement within group from BL2 to T3 ($t(17) = -3.50$, $p < 0.05$) in the SDMT. The performance in the SDMT in group B further increased from T3 to T4 ($t(17) = -3.12$, $p < 0.05$) combined with an increase in the digit span performance ($t(17) = -2.34$, $p < 0.05$). In group C (MSLxWM), the univariate analyses revealed a significant increase in performance in the PASAT ($t(17) = -2.22$, $p < 0.05$) from BL2 to T3 and a significant increase in the SDMT ($t(17) = -5.21$, $p < 0.001$) from T3 to T4. All other univariate comparisons showed no significant results. All means, standard deviations, p-values and Cohens *d* are displayed in table 4.

Table 4. Means, standard deviations and univariate comparisons of the neuropsychological assessment at each session for each group.

	BL2	**T3**	**T4**	**BL2 vs T3**		**T3 vs T4**	
A (WM+MSL)				p	d	p	d
PASAT	52.17±8 0.2	54.39 ± 7.52	55.94 ± 6.66	.088	.28	.144	.22
SDMT	69.17 ± 13.50	76.00 ± 17.59	78.44 ± 15.26	.007*	.39	.099	.01
Corsi Block	17.17 ± 4.25	16.89 ± 4.71	17.33 ± 4.52	.790	.06	.260	.09
Digit Span	16.56 ± 2.83	17.33 ± 3.18	17.61 ± 3.42	.181	.26	.472	.08
9HPT right	16.99 ± 1.78	16.81 ± 1.97	16.06 ± 1.77	.660	.09	.116	.34
9HPT left	17.57 ± 2.48	17.95 ± 2.67	18.05 ± 2.70	.563	.15	.854	.04
B (MSL+WM)							
PASAT	54.22 ± 4.31	52.00 ± 11.94	56.11 ± 5.65	.435	.24	.114	.40
SDMT	68.44 ± 14.61	75.67 ± 17.73	80.83 ± 18.20	.003*	.42	.006*	.29
Corsi Block	16.00 ± 4.39	16.89 ± 4.60	17.39 ± 4.92	.187	.19	.276	.10
Digit Span	15.67 ± 2.93	15.28 ± 3.12	16.56 ± 2.94	.537	.13	.032*	.42
9HPT right	16.72 ± 2.14	16.75 ± 1.69	16.19 ± 1.74	.948	.01	.093	.32
9HPT left	18.07 ± 1.90	18.03 ± 1.83	17.60 ± 1.31	.899	.02	.237	.26
C (WMxMSL)							
PASAT	53.78 ± 7.80	56.39 ± 3.99	-	.041*	.30	-	
SDMT	73.28 ± 17.79	76.50 ± 17.34	-	.127	.18	-	
Corsi Block	16.28 ± 3.56	15.39 ± 4.09	-	.311	.23	-	
Digit Span	16.67 ± 3.05	17.28 ± 3.16	-	.232	.19	-	
9HPT right	17.11 ± 1.89	16.92 ± 2.21	-	.595	.09	-	
9HPT left	17.19 ± 1.92	17.09 ± 2.11	-	.800	.05	-	

Note. For PASAT, SDMT, corsi block and digit span number of correct answers are displayed. 9HPT was measured in seconds. **, significance level at $p < .001$, *, significance level at $p < .05$, d = Cohens' d.

3.2 fMRI results

Interestingly, the FC analysis performed on the whole sample showed no differences between the BL1 and BL2 MRI sessions. The comparisons between the BL1 (first MRI) and the T4 MRI revealed increased FC after the training in brain regions belonging to a parieto-fronto-temporal network and cerebellum, as highlighted by the NBS analysis (figure 4). Within this network, the right inferior parietal gyrus and the left posterior-superior temporal gyrus showed increased connectivity with the majority of the other regions suggesting increased centrality of these regions within this subnetwork (figure 5).

Notably, the right inferior parietal gyrus as defined in the Desikan-Killiany cortical atlas includes the angular gyrus (AG). Specifically, the right inferior parietal gyrus showed increased FC with right middle temporal gyrus, right orbitofrontal gyrus and the right hippocampus. The left posterior-superior temporal gyrus showed increased connectivity with the right superior parietal gyrus, the right supramarginal gyrus, the right postcentral gyrus, the right anterior cingulate, the right insula and right amygdala. Moreover, other relevant regions showing an increased connectivity after the trainings are the left precentral gyrus, the right superior parietal and the middle temporal gyrus bilaterally (table 4).

The subgroup analysis performed using the Cohen's d showed the highest effect size between BL1 and T4 in group B, and between BL1 and T3 in group C. Group B showed a Cohen's d higher than 0.8 among regions belonging to the default mode network (DMN) and the visual network, the sensorimotor and the dorsal attention, the sensorimotor and the frontoparietal and between the limbic and visual network. The group C showed the higher effect size (>0.8) between dorsal attention and DMN and between the sensorimotor and the DMN. Finally, group A showed the highest effect size (>0.5) between the DMN and the dorsal attentional network, the visual network and the cerebellum.

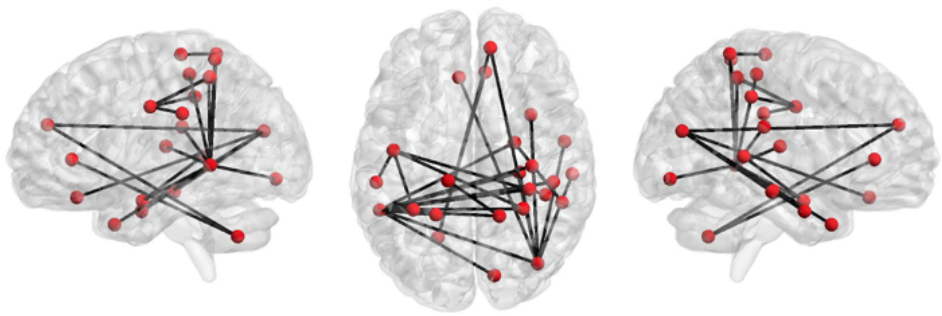

Figure 4. Comparison between session 1 and 4 across the whole sample. The NBS results showed increased connectivity between fronto-parietal regions in the right hemisphere and between left temporal and right parietal regions (Table 4).

Table 4. Connections belonging to the NBS subnetwork showing increased connectivity after training compared to BL1.

Connections		T value
1	right superiorfrontal 2 – right inferiorparietal	4.3
2	right inferiorparietal 6 – right temporalpole 1	4.2
3	right inferiorparietal 6 - right_middletemporal 3	4.5
4	right inferiorparietal 6 - right_middletemporal 4	4.2
5	right inferiorparietal 6 – right hippocampus	4.6
6	right bank-STS 1 – right amygdala.	4.6
7	right inferiorparietal_6 – left medialorbitofrontal 2	4.4
8	right insula 1 – left parsopercularis 2	4.5
9	right hippocampus – left parsopercularis 2	4.2
10	right amygdala – left parsopercularis 2	4.9
11	right superiorparietal 2 – left precentral 1	4.1
12	right supramarginal 1 – left precentral 6.	4.1
13	right superiorparietal 1 – left precentral 6.	4.0
14	left precentral 6 – left supramarginal 2	4.0
15	left precentral 6 – left superiorparietal 2	4.4
16	right inferiorparietal 5 – left middletemporal 4	4.3
17	right inferiorparietal 6 – left middletemporal 4	4.2
18	right postcentral 4 – left bank-STS1	4.1
19	right postcentral 4 – left bank-STS 2	4.2
20	right supramarginal 4 – left bank-STS 2	4.5
21	right superiorparietal 1 – left bank-STS 2	4.4
22	right superiorparietal 2 – left bank-STS 2	5.0
23	right inferiorparietal 6 – left bank-STS 2	4.2
24	right lingual 3 – left bank-STS 2	4.4
25	right insula 1 – left bank-STS 2	4.8
26	right amygdala – bank-STS 2	4.3
27	left superiorparietal 1- bank-STS 2	4.5
28	right superiorfrontal 2 – left lobuleB 7	4.1
29	right rostralanteriorcingulate 1 – left lobuleB 7	4.1

Notes. Numbers next to brain region refers to the Lausanne 2008 atlas.

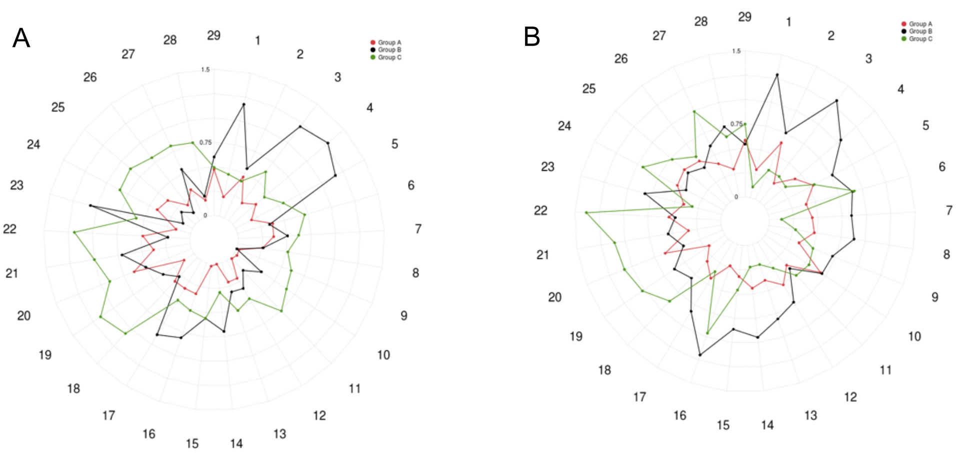

Figure 5. Different patterns of increased connectivity across subgroups assessed computing Cohen's d for all significant connections in the whole sample analysis. The numbers represent the connections displayed in table 4. Red represents group WM+MSL, black represents group MSL+WM and green represents the group WMxMSL. A) session 1 vs session 3; B) session 1 vs session 4.

4. Discussion

In the present study we investigated the interaction between a MSL training and a WM training in healthy subjects using brain FC and neuropsychological outcomes. On the behavioral level, increased processing speed, verbal WM and manual dexterity was found following the trainings. Although the groups did not differ in the training effects, the univariate analyses suggest that MSL conducted before WM training led to additive effects in processing speed and verbal WM. The analyses of the double BLs indicate substantial practice effects in processing speed, verbal WM and manual dexterity but not visuospatial WM. On the neurofunctional level, the analysis on the whole sample showed increased resting state FC after the trainings in a complex network that involved mainly the right angular gyrus (AG), the bank of the left superior temporal sulcus, the right superior parietal gyrus, the middle temporal gyrus bilaterally and the left precentral gyrus. The subgroup analyses showed the highest effect size for increased FC in the mentioned brain regions in the group that performed MSL before WM training, followed by the group that did both trainings combined. It is important to underline that the comparison between the two sessions performed before the trainings did not show any significant difference in FC, pointing to a training-specific effect.

All mentioned brain regions are involved in functions relevant for both MSL and WM trainings. Indeed, the AG is a region located in the portion of the inferior parietal gyrus adjacent to the temporal and occipital lobes which has been described as a continuation of the inferior parietal lobule through the superior and middle temporal gyri (43). Due to its position, the AG is thought to have a critical role in integrating information between multiple input modalities and brain networks (43) and has therefore been suggested to be involved in brain functions such as attention, spatial cognition, memory retrieval, reading, comprehension, number processing (44), visuospatial attention (45), episodic memory (46) but also semantic memory (43,47). Although those cognitive functions express different features, a unifying model has recently been suggested to explain the common engagement of the AG in semantic and episodic memory. The model proposes that the AG combines varying forms of information from multiple sensory modalities or spatiotemporal frameworks as an integrative dynamic buffer (48). The integration of different inputs in the AG may then result in processes such as attention shifting to task-relevant information (43), which could be explained through the participation of the AG in a “bottom-up” attentional subsystem (49). Indeed, studies on FC showed that the AG is part of different networks - most consistently of the DMN, which has been associated with brain activity at rest (50). However, recent literature suggests that the DMN – including the AG - is not only involved in rest situations, but also the unconscious processing of implicit memory (51) and WM tasks (52). Indeed, changes in task-related FC within hubs of the DMN, specifically in the bilateral AG have been described, which were explained through the buffer role of the AG in integrating phonological and visual processes (52). Our study not only supports the role of the AG as an integrative region whose activity could be increased as a result of its involvement in WM and implicit memory processes, it also extends the previous studies in two ways. First, the increased FC in the AG following the trainings in our study was found at rest, which further supports the role of the AG as a DMN hub in the unconscious processing of WM. Second, the increased resting state FC in the AG showed highest effect size in the group that performed the MSL before the WM training, suggesting an involvement of the AG in motor learning.

Indeed, in our study we found an increased resting state FC in the bilateral middle temporal gyri together with the AG following the WM and MSL trainings, specifically pronounced in the MSL+WM group. The AG combined with the bilateral middle temporal gyrus have been described to be involved in action-feedback monitoring following hand movement (53). Specifically, the angular and middle temporal gyri have been shown to be involved in intersensory conflict detection, suggesting a contribution to awareness of temporal discrepancies (53). Furthermore, the middle temporal gyrus has also been identified in a study on healthy adults that described increased activity following simple finger movement (54). The increased activity in the right AG, left middle frontal gyrus, bilateral post-central gyri, superior parietal gyrus and cerebellum could be defined by their role in semantic memory related to voluntary movement. The activation in the left inferior, middle and superior temporal gyri as well as the bilateral inferior frontal gyri seemed to be associated to the ideation of the finger movement and not the movement *per se* (54). Indeed, previous studies described resting state FC changes in the sensorimotor and fronto-parietal networks one hour after motor training, suggesting an offline processing of the newly learned motor skills (19). Similarly, the middle temporal gyrus has been shown to be increased at rest following WM training (18). Thus, the increased resting state FC in the middle temporal gyrus together with the AG in our study suggests an involvement of the middle temporal gyrus in detection and processing functions of finger movement and WM. More importantly, we detected changes in resting state which indicates an offline processing following MSL and WM training, reflecting ongoing learning mechanisms. This notion is further supported by the increased resting state FC shown in the left precentral gyrus post-training which mirrors the participants' performance of all trainings with their dominant right hand. Additionally, resting state FC changes in the precentral gyrus following motor training have been reported previously (19,55) which together with our findings strengthens its involvement in learning processes.

In this context, we also observed an increased FC of the right superior parietal gyrus. A large body of evidence has shown that attentional control involves the parietal cortex, including the intraparietal sulcus and superior parietal gyrus (56). Indeed, the right superior parietal gyrus is a relevant brain region involved in sustained attention, a crucial component of learning and memory (57,58). Additionally, it has also been reported that the bilateral superior parietal gyrus plays an important role in enhancing short-term MSL during observation of hand movements (59). Furthermore, the increased FC in the right superior parietal gyrus found in our study was mainly related to increased FC in the left posterior-superior temporal gyrus and left superior temporal sulcus. This finding is in line with a meta-analytical connectivity model, which described a coactivation between the left superior temporal sulcus and the right superior temporal gyrus, extending to the middle temporal gyrus (60). While the middle temporal gyrus seems to be involved in movement processes and the right superior parietal gyrus has additionally been shown to be involved in to attentional processes, the left superior temporal sulcus has been shown to be involved in the processing of visual movement consequences (61). Overall, the described activation patterns in the superior parietal gyrus, superior temporal sulcus and middle temporal gyrus seem to correspond to the so-called dorsal stream within the dual stream theory of visuospatial processing (62,63). According to the dual stream theory, visual information reaches the parietal lobe through the lateral intraparietal area in order to access the superior temporal sulcus and middle temporal gyrus (64). Thus, the dorsal stream delivers information direct to the motor system for immediate use for reaching, grasping or eye movements (65). Additionally, it has been suggested that the parieto-prefrontal pathway links the middle temporal gyrus, intraparietal areas and the prefrontal cortex, areas which have been suggested to be relevant for spatial WM (64). By contrast, the ventral stream describing temporal regions is dedicated to 'vision-for-perception', but has further been described to have a role in movement planning based on memory of the object and its relationship to other items (64,65). Despite the distinction in dorsal and ventral streams, it has been suggested that during hand movement both streams are cross-communicating through the temporo-parietal fibers (66). The increase connectivity of the AG with other regions (middle temporal gyrus, right superior parietal gyrus, left superior temporal sulcus, left precentral gyrus) could suggest that the AG may act as an integrative region that could promote the cross-communication between dorsal and ventral streams. Additionally, they extend this notion since we observed increased FC within and between both streams in the resting brain following WM and MSL training, which can be seen as a cross-communication in terms of an offline processing.

Moreover, our results allow to understand the direction of the cross-communication, since the sequential order of the training administration modulated FC changes in the previously mentioned networks. Indeed, the post-training resting state FC changes showed the highest effect size in the group which completed the MSL first followed by the WM training and the group which did both trainings combined. Current research suggests that the motor cognitive interdependence may come from motor systems that dedicate neuronal regions to cognitive demands, which results in enhancement of active rehearsal processes through internally generated motor sequence traces that are actively recreated at will (15). Behaviorally, we detected additive effects in visuospatial WM and processing speed following the group which first completed the MSL before the WM training. Hence, MSL training before or simultaneously with WM training seems to engage motor and cognitive brain regions which could act as a boost for WM and processing speed ability. It is important to underline that since all FC changes were detected in the brain at rest, the modulations due to the sequential order of the training seems to reflect more pronounced offline processing when the MSL is done before the WM training. The absence of FC differences between the BL1 and BL2 further supports that the observed changes in resting state FC are a result of the trainings.

Considering the relatively young age of the participants (average age 30 years) the generalization of our findings to an older population should be done with caution. Indeed, it has been described that resting state FC patterns between young and old adults differ following MSL (19). Similarly, a recent study showed increased resting state FC following a short-term motor practice in young adults, whereas decreased FC was found in older adults (55). For this reason, the results in this study cannot be generalized and future studies should investigate resting state FC changes following MSL and WM training in other age population, specifically old age.

5. Conclusion

To our best knowledge this is the first study that investigated the brain mechanisms at rest related to additive and interactive effects of MSL and WM trainings. The results showed distinct patterns of resting state FC modulation related to sequential and combined training programs suggesting a relevance of the order in which trainings are performed. These training-related FC modulations are supported by the absence of differences between the two pre-training sessions. Thus, the current study sheds light on the additive and interactive neuroplastic mechanisms induced by motor and cognitive trainings. Based on this observation, we think that rehabilitative programs may consider to take into account the sequential order of training administration, although further studies in pathological conditions are essential.

6. Acknowledgment

We want to thank all participants for their participation and the University of Basel for supporting this project. Part of this work, namely the use of COGNI-TRAcK, has been performed under formal agreement between the Department of Neurology, University Hospital Basel, University of Basel (Switzerland) and the Italian Multiple Sclerosis Foundation, Genoa (Italy) that supported COGNI-TRAcK design and validation (project n. 2011/R/8).

7. Funding

This work was supported by the University of Basel [grant number DMS2310].

8. References

1. Kitago T, Krakauer JW. Motor learning principles for neurorehabilitation. In: Handbook of Clinical Neurology. 2013. p. 93–103.
2. Willingham DB. A Neuropsychological Theory of Motor Skill Learning. Psychol Rev. 1998;105(3):558–84.
3. Maxwell JP, Masters RSW, Eves FF. The role of working memory in motor learning and performance. Conscious Cogn. 2003;12(3):376–402.
4. Boyd LA, Winstein CJ. Implicit motor-sequence learning in humans following unilateral stroke: The impact of practice and explicit knowledge. Neurosci Lett. 2001;298(1):65–9.
5. Anguera JA, Reuter-Lorenz PA, Willingham DT, Seidler RD. Contributions of spatial working memory to visuomotor learning. J Cogn Neurosci. 2010;22(9):1917–30.
6. Baddeley A. Working memory: Looking back and looking forward. Nat Rev Neurosci. 2003;4(10):829–39.
7. Baddeley A. Working Memory: Theories, Models, and Controversies. Annu Rev Psychol. 2012;63(1):1–29.
8. Diamond A. Executive Functions. Annu Rev Psychol. 2013;64:135–68.
9. Glisky E. Changes in Cognitive Function in Human Aging. In: books.google.com. 2007. p. 3–20.
10. Klingberg T. Training and plasticity of working memory. Vol. 14, Trends in Cognitive Sciences. 2010. p. 317–24.
11. Jaeggi S, Buschkuehl M, Jonides J, Perrig WJ. Improving fluid intelligence with training on working memory. Proc Natl Acad Sci U S A. 2008;105(19):6829–33.
12. Klingberg T, Fernell E, Olesen PJ, Johnson M, Gustafsson P, Dahlström K, et al. Computerized training of working memory in children with ADHD - A randomized, controlled trial. J Am Acad Child Adolesc Psychiatry. 2005;44(2):177–86.
13. Seidler RD, Bo J, Anguera JA. Neurocognitive contributions to motor skill learning: The role of working memory. Vol. 44, Journal of Motor Behavior. Taylor & Francis Group; 2012. p. 445–53.
14. Bo J, Jennett S, Seidler RD. Working memory capacity correlates with implicit serial reaction time task performance. Exp Brain Res. 2011;214(1):73–81.
15. Marvel CL, Morgan OP, Kronemer SI. How the motor system integrates with working memory. Vol. 102, Neuroscience and Biobehavioral Reviews. Elsevier Ltd; 2019. p. 184–94.
16. Coynel D, Marrelec G, Perlbarg V, Pélégrini-Issac M, Van de Moortele PF, Ugurbil K, et al. Dynamics of motor-related functional integration during motor sequence learning. Neuroimage. 2010 Jan 1;49(1):759–66.
17. Salmi J, Nyberg L, Laine M. Working memory training mostly engages general-purpose large-scale networks for learning. Vol. 93, Neuroscience and Biobehavioral Reviews. Elsevier Ltd; 2018. p. 108–22.
18. Jolles DD, Van Buchem MA, Crone EA, Rombouts SARB. Functional brain connectivity at rest changes after working memory training. Hum Brain Mapp. 2013;34(2):396–406.
19. Mary A, Wens V, Op de Beeck M, Leproult R, De Tiège X, Peigneux P. Age-related

differences in practice-dependent resting-state functional connectivity related to motor sequence learning. Hum Brain Mapp. 2017;38(2):923–37.
20. Oldfield RC. The assessment and analysis of handedness: The Edinburgh inventory. Neuropsychologia. 1971;9(1):97–113.
21. Tacchino A, Pedullà L, Bonzano L, Vassallo C, Battaglia MA, Mancardi G, et al. A New App for At-Home Cognitive Training: Description and Pilot Testing on Patients with Multiple Sclerosis. JMIR mHealth uHealth. 2015;3(3):e85.
22. Pedullà L, Brichetto G, Tacchino A, Vassallo C, Zaratin P, Battaglia MA, et al. Adaptive vs. non-adaptive cognitive training by means of a personalized App: a randomized trial in people with multiple sclerosis. J Neuroeng Rehabil. 2016;13(1):88.
23. Bonzano L, Pedullà L, Pardini M, Tacchino A, Zaratin P, Battaglia MA, et al. Brain activity pattern changes after adaptive working memory training in multiple sclerosis. Brain Imaging Behav. 2020;14(1):142–54.
24. Gronwall DMA. Paced auditory serial addition task: A measure of recovery from concussion. Percept Mot Skills. 1977 Apr;44(2):367–73.
25. Smith A. Symbol digit modalities test. Western Psychological Services., editor. Los Angeles, CA; 1982. 22 p.
26. Wechsler D. Wechsler Adult Intelligence Scale–Fourth Edition (WAIS–IV). San Antonio, TX: The Psychological Corporation; 2008.
27. Corsi P. Memory and the medial temporal region of the brain. McGill University. Montreal, QB.; 1972.
28. Mathiowetz V, Weber K, Kashman N, Volland G. Adult norms for the nine hole peg test of finger dexterity. Occup Ther J Res. 1985;5(1):24–38.
29. Xu J, Moeller S, Auerbach EJ, Strupp J, Smith SM, Feinberg DA, et al. Evaluation of slice accelerations using multiband echo planar imaging at 3T. Neuroimage. 2013 Dec;83:991–1001.
30. Glasser MF, Sotiropoulos SN, Wilson JA, Coalson TS, Fischl B, Andersson JL, et al. The minimal preprocessing pipelines for the Human Connectome Project. Neuroimage. 2013 Oct;80:105–24.
31. Fischl B, Dale AM. Measuring the thickness of the human cerebral cortex from magnetic resonance images. Proc Natl Acad Sci U S A. 2000 Sep;97(20):11050–5.
32. Destrieux C, Fischl B, Dale A, Halgren E. Automatic parcellation of human cortical gyri and sulci using standard anatomical nomenclature. Neuroimage. 2010;53(1):1–15.
33. Fischl B, Salat DH, Busa E, Albert M, Dieterich M, Haselgrove C, et al. Whole Brain Segmentation: Automated Labeling of Neuroanatomical Structures in the Human Brain. Neuron. 2002;33:341–55.
34. Daducci A, Gerhard S, Griffa A, Lemkaddem A, Cammoun L, Gigandet X, et al. The Connectome Mapper: An Open-Source Processing Pipeline to Map Connectomes with MRI. PLoS One. 2012;7(12).
35. Weier K, Fonov V, Lavoie K, Doyon J, Louis Collins D. Rapid automatic segmentation of the human cerebellum and its lobules (RASCAL)-Implementation and application of the patch-based label-fusion technique with a template library to segment the human cerebellum. Hum Brain Mapp. 2014;35(10):5026–39.
36. Hagmann P, Cammoun L, Gigandet X, Meuli R, Honey CJ, Wedeen VJ, et al. Mapping the Structural Core of Human Cerebral Cortex. Friston KJ, editor. PLoS Biol. 2008 Jul;6(7):e159.
37. Smith SM, Jenkinson M, Woolrich MW, Beckmann CF, Behrens TEJ, Johansen-Berg H, et al. Advances in functional and structural MR image analysis and implementation as FSL. In: NeuroImage. 2004. p. S208-S219.
38. Andersson JLR, Skare S, Ashburner J. How to correct susceptibility distortions in spin-echo echo-planar images: application to diffusion tensor imaging. Neuroimage. 2003 Oct;20(2):870–88.
39. Yeo BTT, Krienen FM, Sepulcre J, Sabuncu MR, Lashkari D, Hollinshead M, et al. The organization of the human cerebral cortex estimated by intrinsic functional connectivity. J Neurophysiol. 2011;106:1125–65.

40. R Core Team. R: A language and environment for statistical computing. R Found Stat Comput. 2013;Vienna(Austria).
41. Zalesky A, Fornito A, Bullmore ET. Network-based statistic: Identifying differences in brain networks. Neuroimage. 2010 Dec;53(4):1197–207.
42. Rubinov M, Sporns O. Complex network measures of brain connectivity: Uses and interpretations. Neuroimage. 2010;52(3):1059–69.
43. Seghier ML. The angular gyrus: Multiple functions and multiple subdivisions. Neuroscientist. 2013;19(1):43–61.
44. Brannon EM. The representation of numerical magnitude. Curr Opin Neurobiol. 2006;16(2):222–9.
45. Studer B, Cen D, Walsh V. The angular gyrus and visuospatial attention in decision-making under risk. Neuroimage. 2014;103:75–80.
46. Thakral PP, Madore KP, Schacter DL. A role for the left angular gyrus in episodic simulation and memory. J Neurosci. 2017;37(34):8142–9.
47. Binder JR, Desai RH. The neurobiology of semantic memory. Vol. 15, Trends in Cognitive Sciences. 2011. p. 527–36.
48. Humphreys GF, Ralph MAL, Simons JS. A Unifying Account of Angular Gyrus Contributions to Episodic and Semantic Cognition. 2020;1–21.
49. Ciaramelli E, Grady CL, Moscovitch M. Top-down and bottom-up attention to memory: A hypothesis (AtoM) on the role of the posterior parietal cortex in memory retrieval. Vol. 46, Neuropsychologia. 2008. p. 1828–51.
50. Buckner RL, Andrews-Hanna JR, Schacter DL. The brain's default network: Anatomy, function, and relevance to disease. Vol. 1124, Annals of the New York Academy of Sciences. John Wiley & Sons, Ltd; 2008. p. 1–38.
51. Yang J, Weng X, Zang Y, Xu M, Xu X. Sustained activity within the default mode network during an implicit memory task. Cortex. 2010;46(3):354–66.
52. Vatansever D, Manktelow AE, Sahakian BJ, Menon DK, Stamatakis EA. Angular default mode network connectivity across working memory load. Hum Brain Mapp. 2017;38(1):41–52.
53. Van Kemenade BM, Arikan BE, Podranski K, Steinsträter O, Kircher T, Straube B. Distinct Roles for the Cerebellum, Angular Gyrus, and Middle Temporal Gyrus in Action-Feedback Monitoring. Cereb Cortex. 2019;29(4):1520–31.
54. Caffarra P, Gardini S, Vezzadini G, Bromiley A, Venneri A. The ideation of movement is supported by fronto-temporal cortical regions involved in the retrieval of semantic knowledge. Acta Biomed l'Ateneo Parm. 2010;81(1):21–9.
55. Solesio-Jofre E, Beets IAM, Woolley DG, Pauwels L, Chalavi S, Mantini D, et al. Age-dependent modulations of resting state connectivity following motor practice. Front Aging Neurosci. 2018;10(FEB):25.
56. Thakral PP, Slotnick SD. The role of parietal cortex during sustained visual spatial attention. Brain Res. 2009;1302:157–66.
57. Cowan N. Attention and Memory: An Integrated Framework. Attention and Memory: An Integrated Framework. 2008. 1–336 p.
58. Sarter M, Givens B, Bruno JP. The cognitive neuroscience of sustained attention: Where top-down meets bottom-up. Vol. 35, Brain Research Reviews. 2001. p. 146–60.
59. Ossmy O, Mukamel R. Activity in superior parietal cortex during training by observation predicts asymmetric learning levels across hands. Sci Rep. 2016;6(February):1–9.
60. Erickson LC, Rauschecker JP, Turkeltaub PE. Meta-analytic connectivity modeling of the human superior temporal sulcus. Brain Struct Funct. 2017;222(1):267–85.
61. Limanowski J, Sarasso P, Blankenburg F. Different responses of the right superior temporal sulcus to visual movement feedback during self-generated vs. externally generated hand movements. Eur J Neurosci. 2018;47(4):314–20.
62. Mishkin M, Ungerleider LG. Two cortical visual systems. Anal Vis Behav. 1982;549–586.

63. Rizzolatti G, Matelli M. Two different streams form the dorsal visual system: anatomy and functions. Exp Brain Res. 2003;153:146–57.
64. Kravitz DJ, Saleem KS, Baker CI, Mishkin M. A new neural framework for visuospatial processing. 2011;
65. van Polanen V, Davare M. Interactions between dorsal and ventral streams for controlling skilled grasp. Neuropsychologia. 2015;79:186–91.
66. Budisavljevic S, Dell'Acqua F, Castiello U. Cross-talk connections underlying dorsal and ventral stream integration during hand actions. Cortex. 2018;103:224–39.

CRediT author statement

Priska Zuber: Formal Analysis, Investigation, Data Curation, Writing – Original Draft, Visualization, Writing - Review & Editing; **Laura Gaetano**: Software, Methodology, Writing - Review & Editing; **Alessandra Griffa**: Methodology, Writing - Review & Editing; **Manuel Huerbin**: Investigation, Writing - Review & Editing; **Ludovico Pedulla** & **Laura Bonzano**: Software, Writing - Review & Editing; **Anna Altermatt** & **Charidimos Tsagkas**: Investigation, Writing - Review & Editing; **Katrin Parmar**: Software, Writing - Review & Editing; **Patric Hagmann**: Methodology, Formal Analysis, Writing - Review & Editing; **Jens Wuerfel**: Resources, Wrting – Review & Editing; **Ludwig Kappos** & **Till Sprenger**: Resources, Writing - Review & Editing; **Olaf Sporns**: Formal Analysis, Writing - Review & Editing, Resources; **Stefano Magon**: Conceptualization, Methodology, Formal Analysis, Visualization, Supervision, Project Administration, Funding Acquisition, Writing – Original Draft, Writing - Review & Editing.

6 Discussion

Although brain plasticity is a central concept for both recovery from brain damage in pathology and performance enhancement in health, insufficient investigation of underlying neural, cognitive or motor mechanisms limits the understanding of the extent of improvement on various functional domains. Three experimental studies conducted in the frame of this thesis aimed at studying neural and cognitive plasticity induced by interventions in order to understand their mechanisms at action from different perspectives. In study **A)**, we investigated the neural and behavioral effects of a personalized, multidisciplinary rehabilitation program in MS using fMRI. Study **B)** targeted current challenges in cognitive training research by studying the comparative efficacy and task-related cognitive mechanisms of a model-based WM training in healthy old adults. In study **C)**, additive and interactive effects of WM and motor training were investigated using fMRI in healthy young adults. All studies indicate plastic changes following the administered interventions on different levels.

The results of the first study indicate improved self-perceived fatigue, walking ability and health-related quality of life following an inpatient personalized multidisciplinary rehabilitation program in people with MS. Additionally, participants showed improved accuracy on an untrained MSL task after the rehabilitation. On the neural level, task-fMRI indicated a reduced difference between the random and repeated condition of a MSL task in the left cerebellum and right prefrontal lobe after the rehabilitation in people with MS compared to healthy controls. According to neuroplasticity in the sense of map expansion, a decrease in activation may be the result of an initial task-related activation of the whole network until the representational region is selected (Grafman, 2000). Indeed, in cross-sectional studies it has been shown that people with MS display increased motor-task related activation in sensorimotor areas and cerebellum compared to healthy controls (De Giglio et al., 2016). Longitudinally, increased or decreased activation patterns following motor training have been reported (Dayan, E., & Cohen, 2011; De Giglio et al., 2018; Floyer-Lea & Matthews, 2005). The decreased activation following the rehabilitation in our study thus suggests a more effective recruitment of cerebellar and prefrontal regions in the sense of map expansion in people with MS. In extension to these previous findings, this study is amongst the first to describe clinical and neurofunctional plasticity following a clinically defined inpatient personalized multidisciplinary rehabilitation. Thus, our study supports activity-induced neuroplasticity as an interface to recovery (Horton et al., 2017) in the sense of a personalized multidisciplinary rehabilitation, a setting aiming at reducing the disease-related burden by taking the polysymptomatic presentation of MS into consideration.

The results of this first study are limited in the sense that the described neuroplastic changes cannot be generalized to cognitive functions. This is related to the fact that the investigated inpatient multidisciplinary rehabilitation focused on motor functions. After reviewing the patient-specific rehabilitative therapies in our study, 1±1.7 hours were dedicated to cognitive rehabilitation of memory, attention and executive functions compared to on average 32.8±8.6 hours dedicated to physiotherapy. Interestingly, this observation diverges from the literature, which shows that cognitive rehabilitation is broadly investigated and indicates positive effects on cognitive functions in people with MS (Mitolo, Venneri, Wilkinson, & Sharrack, 2015). Hence, this observation indicates a mismatch between research on cognitive rehabilitation and its actual translation into the clinical setting. First, this mismatch may be explained in the inconclusive and conflicting evidence regarding the efficacy of cognitive rehabilitation to improve cognition in patients with MS, which can be attributed to the variability of rehabilitative approaches as well as methodological considerations (Mitolo et al., 2015). In light of the supply-demand model of plasticity, variable input is required in order to increase the mismatch between supply and demand and thus induce plasticity. However, the variability needs to be understood in the context of using multiple tasks that target one single cognitive process (Lövdén et al., 2010). Indeed, the current focus of cognitive rehabilitation programs for MS lays in targeting executive control processes or WM (Mitolo et al., 2015).

A second explanation may be found in the current challenges of cognitive training in healthy adults. In order to understand how plasticity can be induced with cognitive rehabilitation programs, underlying mechanisms of cognitive interventions need further clarification and studies on their comparative efficacy are required. On this ground, developing theory-driven training interventions that are first tested in healthy people is unavoidable in order to understand the efficacy and underlying mechanisms which may then be translated into the clinical setting. The two studies conducted within this thesis may have the potential to give insight in challenges that occur in the investigation of cognitive trainings, which may form the basis for cognitive rehabilitation strategies. We first aimed at an in-depth investigation of the mechanism of action as well as the comparative efficacy of a newly developed model-based WM training in healthy older adults (**B**) and second targeted the additive and interactive neural and behavioral effects of WM and motor training in healthy young adults (**C**).

With study **B)**, we addressed two main challenges in cognitive training research simultaneously. We developed a theory-driven WM training based on the multicomponent model (Baddeley, 2012) and experimentally manipulated the inclusion of distractor inhibition as a possible task-related process. This approach allows to draw conclusion on the trained cognitive construct and to investigate if task-related process may render the improvement on the trained cognitive function (Katz et al., 2017; Morrison & Chein, 2011). Further, we tested the comparative efficacy of the model-based WM trainings with and without distractor inhibition to a dual-n-back training (Jaeggi et al., 2008) and an active control group in a parallel group, double-baseline, randomized, controlled clinical. Comparing the trainings further allowed to investigate if potential transfer is paradigm specific. The results revealed improved WM capacity in healthy old adults only following the model-based WM training with distractor inhibition, specifically in comparison to the same training without distractor inhibition. Although no far transfer effects were detected following the completion of all trainings, small near transfer of the dual-n-back paradigm to an untrained n-back task as well as to visuospatial learning following the model-based training with distractor inhibition was found. While the results regarding the n-back training indicate paradigm-specific improvements (Holmes et al., 2019), the distractor inhibition within a model-based WM training indicates a relevant task-related process of WM with the possibility to improve performance on untrained WM capacity tasks in healthy old adults.

Despite the limitations due to early termination of the study, the double baseline approach allows to exclude that the described training gains were primarily based on practice effects and to attribute the detected near transfer post-training to the training itself. Thus, the model-based WM training with distractor inhibition has the potential to increase the supply-demand mismatch required to successfully induce cognitive plasticity (Lövdén et al., 2010). Although no far transfer was detected, the near transfer to WM capacity could not only be relevant in healthy participants, but also for a clinical population. The model-based approach offers one training task for each WM component separately which enables the possibility to train the specific needs of patients based on their individual WM impairment by selecting the appropriate task. While the results cannot be generalized to a clinical population yet, testing the model-based WM training in clinical conditions such as MS in future studies could display the basis of an effective, evidence-based personalized cognitive rehabilitation approach for WM.

The last study presented within this thesis (**C**) showed increased resting state FC in the right angular gyrus, the bank of the left superior temporal sulcus, the right superior parietal gyrus, the middle temporal gyrus bilaterally and the left precentral gyrus after a WM and motor training in healthy young adults. The resting state FC in those regions was modulated by the order in which trainings were performed, with highest effect sizes in the group that performed motor before WM training, followed by the group that did both trainings combined. On the behavioral level, results showed increased processing speed and verbal WM following the trainings, specifically in the group that performed motor before WM training. While supporting the previously suggested shared neural and behavioral mechanisms between WM and motor learning (Marvel et al., 2019), these results are the first to reveal a relevance of the order in which motor and cognitive training is performed on the resulting cognitive and neural plasticity. We brought together two previous observations. First, it has been suggested that offline processing of newly learned motor skills as resting state FC changes in sensorimotor and frontoparietal networks takes place after motor training (Mary et al., 2017). Second, the motor-cognitive interdependence has been described to result in active rehearsal processes through internally generated motor sequence traces that are actively recreated at will (Marvel et al., 2019). Thus, completing motor training before or combined with WM training may act as a boost for WM and processing speed since offline processing of motor skills may induce motor sequence traces that could facilitate active rehearsal of the WM input.

This study is limited due to its low generalizability to other study populations, since resting state FC patterns following motor training may differ in older adults or because of pathological conditions (De Giglio et al., 2018; Solesio-Jofre et al., 2018). Nevertheless, the high flexibility of the human motor system and its continuous interaction with cognitive processes (Horton et al., 2017) combined with our findings highlights the potential of the motor-cognitive interaction for recovery after brain damage. The administration of a motor training before or in combination with cognitive rehabilitation may be more successful in improving cognitive impairment in patients than completing cognitive rehabilitation alone. Thus, the role of sequential order of WM and motor training administration gives valuable insight on neuroplastic changes, which may be crucial in the planning and designing of effective rehabilitative programs for people with pathological conditions.

In summary, the three studies presented within this thesis give insight on brain plasticity in health and pathology from various perspectives by applying behavioral and functional imaging methods. Following the current focus in rehabilitation and cognitive enhancement research, the importance of comprehensive examination of underlying neural and cognitive mechanisms of intervention induced plasticity was demonstrated. We showed that inpatient personalized multidisciplinary rehabilitation program in people with MS induces not only improvement in highly impacting symptoms but also neuroplastic changes. Further, we identified distractor inhibition as a task-related process of WM training in order to induce cognitive plasticity in healthy older adults and revealed neural and behavioral effects related to the order in which motor and cognitive trainings are performed in healthy young adults. Our findings in the healthy population provide a starting point for effective cognitive trainings that could eventually be translated into rehabilitative strategies addressing individual WM impairment. Indeed, cognitive impairment is amongst the symptoms occurring in the first year after disease onset in patients with MS (Kister et al., 2013), which highlights the importance of rehabilitative strategies to improve cognition in patients early on. Due to the polysymptomatic clinical presentation of MS, it has been argued that therapeutic interventions should be developed in order to allow a personalized rehabilitative care program (Amatya et al., 2019). Future studies should therefore evaluate if the model-based WM tasks may improve WM capacity in a clinical sample and if distractor inhibition further displays a task-relevant process for WM improvement in patients suffering from WM impairment. Based on the previously described differences of brain activity between healthy adults and patients with MS (De Giglio et al., 2018), it would be interesting to see if the modulation in brain activity according to the order of motor and cognitive training administration can further be replicated in a clinical sample.

In conclusion, it is demonstrated that brain plasticity following interventions in health and pathology is the result of an interplay between various behavioral and functional mechanisms. Identifying processes of cognitive trainings based on well-established theoretical models answers if and more importantly how transfer effects can occur. Furthermore, future empirical studies should continue to evaluate additive and interactive effects of different functional domains and their potential in inducing brain plasticity in healthy people. Such studies form the basis for developing successful rehabilitative strategies and have the power to bridge between basic neuroscientific and interventional research towards the translation into the clinical setting.

7 References

Amatya, B., Khan, F., & Galea, M. (2019). Rehabilitation for people with multiple sclerosis: An overview of Cochrane Reviews. *Cochrane Database of Systematic Reviews*, Vol. 2019. https://doi.org/10.1002/14651858.CD012732.pub2

Baddeley, A. (2003). Working memory: Looking back and looking forward. *Nature Reviews Neuroscience*, *4*(10), 829–839.

Baddeley, A. (2012). Working Memory: Theories, Models, and Controversies. *Annual Review of Psychology*, *63*(1), 1–29.

Baddeley, A., Hitch, G., & Bower, G. H. (1974). *Working memory. The psychology of learning and motivation*. 47–89.

Basak, C., Qin, S., & O'Connell, M. A. (2020). Differential Effects of Cognitive Training Modules in Healthy Aging and Mild Cognitive Impairment: A Comprehensive Meta-Analysis of Randomized Controlled Trials. *Psychology and Aging*, *35*(2), 220–249.

Beer, S., Khan, F., & Kesselring, J. (2012). Rehabilitation interventions in multiple sclerosis: An overview. *Journal of Neurology*, *259*(9), 1994–2008.

Borella, E., Ghisletta, P., & De Ribaupierre, A. (2011). Age differences in text processing: The role of working memory, inhibition, and processing speed. *Journals of Gerontology - Series B Psychological Sciences and Social Sciences*, *66 B*(3), 311–320.

Budson, A. E., & Price, B. H. (2007). Memory dysfunction in neurological practice. *Practical Neurology*, *7*(1), 42–47.

Cajal, S. R. y. (1928). *Degeneration and regeneration of the nervous system.*

Chen, J. E., & Glover, G. H. (2015). Functional Magnetic Resonance Imaging Methods. *Neuropsychology Review*, Vol. 25, pp. 289–313. Springer New York LLC.

Cramer, S. C., Sur, M., Dobkin, B. H., O'Brien, C., Sanger, T. D., Trojanowski, J. Q., … Vinogradov, S. (2011). Harnessing neuroplasticity for clinical applications. *Brain*, Vol. 134, pp. 1591–1609. Oxford Academic.

Damoiseaux, J. S., Rombouts, S. A. R. B., Barkhof, F., Scheltens, P., Stam, C. J., Smith, S. M., & Beckmann, C. F. (2006). Consistent resting-state networks across healthy subjects. *Proceedings of the National Academy of Sciences of the United States of America*, *103*(37), 13848–13853.

Dayan, E., & Cohen, L. G. (2011). Neuroplasticity subserving motor skill learning. *Neuron*, *72*(3), 443–454.

De Giglio, L., Tommasin, S., Petsas, N., & Pantano, P. (2018). The role of fMRI in the assessment of neuroplasticity in MS: A systematic review. *Neural Plasticity*, Vol. 2018.

De Giglio, L., Tona, F., De Luca, F., Petsas, N., Prosperini, L., Bianchi, V., … Pantano, P. (2016). Multiple Sclerosis: Changes in Thalamic Resting-State Functional Connectivity Induced by a Home-based Cognitive Rehabilitation Program. *Radiology*, *280*(1), 202–211.

Deroost, N., Smetcoren, S., Vandenbossche, J., & Hooghe, M. (2014). Implicit and Explicit Learning of Sequential Motor Skill in Multiple Sclerosis: Directions for Rehabilitation. *Journal of Neuroscience and Rehabilitation*, *1*(1).

Diamond, A. (2013). Executive Functions. *Annual Review of Psychology*, *64*, 135–168.

Eriksson, J., Vogel, E. K., Lansner, A., Bergström, F., & Nyberg, L. (2015). Neurocognitive Architecture of Working Memory. *Neuron*, Vol. 88, pp. 33–46. Cell Press.

Feinstein, A., Freeman, J., & Lo, A. C. (2015). Treatment of progressive multiple sclerosis: What works, what does not, and what is needed. *The Lancet Neurology*, Vol. 14, pp. 194–207. Lancet Publishing Group.

Floyer-Lea, A., & Matthews, P. M. (2005). Distinguishable Brain Activation Networks for Short- and Long-Term Motor Skill Learning. *Journal of Neurophysiology*, *94*(1), 512–518.

Gathercole, S. E., Dunning, D. L., Holmes, J., & Norris, D. (2019). Working memory training involves learning new skills. *Journal of Memory and Language*, *105*, 19–42. https://doi.org/10.1016/j.jml.2018.10.003

Gazzaley, A., Clapp, W., Kelley, J., McEvoy, K., Knight, R. T., & D'Esposito, M. (2008). Age-related top-down suppression deficit in the early stages of cortical visual memory processing. *Proceedings of the National Academy of Sciences of the United States of America*, *105*(35), 13122–13126.

Glover, G. H. (2011). Overview of functional magnetic resonance imaging. *Neurosurgery Clinics of North America*, Vol. 22, pp. 133–139. https://doi.org/10.1016/j.nec.2010.11.001

Grafman, J. (2000). Conceptualizing functional neuroplasticity. *Journal of Communication Disorders*, *33*(4), 345–356.

Green, C. S., & Bavelier, D. (2008). Exercising Your Brain: A Review of Human Brain Plasticity and Training-Induced Learning. *Psychology and Aging*, *23*(4), 692–701.

Hartshorne, J. K., & Germine, L. T. (2015). When Does Cognitive Functioning Peak? The Asynchronous Rise and Fall of Different Cognitive Abilities Across the Life Span. *Psychological Science*, *26*(4), 433–443. https://doi.org/10.1177/0956797614567339

Holmes, J., Woolgar, F., Hampshire, A., & Gathercole, S. E. (2019). Are Working Memory Training Effects Paradigm-Specific? *Frontiers in Psychology*, *10*. https://doi.org/10.3389/fpsyg.2019.01103

Horton, J. C., Fahle, M., Mulder, T., & Trauzettel-Klosinski, S. (2017). Adaptation, perceptual learning, and plasticity of brain functions. *Graefe's Archive for Clinical and Experimental Ophthalmology*, Vol. 255, pp. 435–447. https://doi.org/10.1007/s00417-016-3580-y

Jaeggi, S., Buschkuehl, M., Jonides, J., & Perrig, W. J. (2008). Improving fluid intelligence with training on working memory. *Proceedings of the National Academy of Sciences of the United States of America*, *105*(19), 6829–6833.

Kamm, C. P., Uitdehaag, B. M., & Polman, C. H. (2014). Multiple sclerosis: Current knowledge and future outlook. *European Neurology*, *72*(3–4), 132–141. https://doi.org/10.1159/000360528

Katz, B., Shah, P., & Meyer, D. E. (2017). How to play 20 questions with nature and lose: Reflections on 100 years of brain-training research. *National Acad Sciences*. https://doi.org/10.1073/pnas.1617102114

Khan, F., & Amatya, B. (2017). Rehabilitation in Multiple Sclerosis: A Systematic Review of Systematic Reviews. *Archives of Physical Medicine and Rehabilitation*, *98*(2), 353–367. https://doi.org/10.1016/j.apmr.2016.04.016

Khan, F., Amatya, B., Galea, M. P., Gonzenbach, R., & Kesselring, J. (2017). Neurorehabilitation: applied neuroplasticity. *Journal of Neurology*, *264*(3), 603–615. https://doi.org/10.1007/s00415-016-8307-9

Kister, I., Bacon, T. E., Chamot, E., Salter, A. R., Cutter, G. R., Kalina, J. T., & Herbert, J. (2013). Natural History of Multiple Sclerosis Symptoms. *International Journal of MS Care*, *15*(3), 146–156. https://doi.org/10.7224/1537-2073.2012-053

Kleim, J. A. (2011). Neural plasticity and neurorehabilitation: Teaching the new brain old tricks. *Journal of Communication Disorders*, *44*(5), 521–528. https://doi.org/10.1016/j.jcomdis.2011.04.006

Klingberg, T. (2010, July). Training and plasticity of working memory. *Trends in Cognitive Sciences*, Vol. 14, pp. 317–324. https://doi.org/10.1016/j.tics.2010.05.002

Klingberg, T., Fernell, E., Olesen, P. J., Johnson, M., Gustafsson, P., Dahlström, K., … Westerberg, H. (2005). Computerized training of working memory in children with ADHD - A randomized, controlled trial. *Journal of the American Academy of Child and Adolescent Psychiatry*, *44*(2), 177–186.

Kloos, A., Gomes-Osman, J., & Boyd, L. (2020). Harnessing Neuroplasticity for Functional Recovery. *Journal of Neurologic Physical Therapy*, Vol. 44, pp. 119–120. https://doi.org/10.1097/NPT.0000000000000307

Kühn, S., & Lindenberger, U. (2015). Research on Human Plasticity in Adulthood: A Lifespan Agenda. In *Handbook of the Psychology of Aging: Eighth Edition* (pp. 105–123). https://doi.org/10.1016/B978-0-12-411469-2.00006-6

Li, C. H., He, X., Wang, Y. J., Hu, Z., & Guo, C. Y. (2017). Visual working memory capacity can be increased by training on distractor filtering efficiency. *Frontiers in Psychology*, *8*(FEB). https://doi.org/10.3389/fpsyg.2017.00196

Lipp, I., & Tomassini, V. (2015). Neuroplasticity and motor rehabilitation in multiple sclerosis. *Frontiers in Neurology*, *6*(MAR). https://doi.org/10.3389/fneur.2015.00059

Lövdén, M., Bäckman, L., Lindenberger, U., Schaefer, S., & Schmiedek, F. (2010). A Theoretical Framework for the Study of Adult Cognitive Plasticity. *Psychological Bulletin*, *136*(4), 659–676. https://doi.org/10.1037/a0020080

Lublin, F. D., Reingold, S. C., Cohen, J. A., Cutter, G. R., Sørensen, P. S., Thompson, A. J., … Polman, C. H. (2014). Defining the clinical course of multiple sclerosis: The 2013 revisions. *Neurology*, Vol.

83, pp. 278–286. https://doi.org/10.1212/WNL.0000000000000560

Marvel, C. L., Morgan, O. P., & Kronemer, S. I. (2019). How the motor system integrates with working memory. *Neuroscience and Biobehavioral Reviews*, Vol. 102, pp. 184–194. Elsevier Ltd.

Mary, A., Wens, V., Op de Beeck, M., Leproult, R., De Tiège, X., & Peigneux, P. (2017). Age-related differences in practice-dependent resting-state functional connectivity related to motor sequence learning. *Human Brain Mapping*, *38*(2), 923–937. https://doi.org/10.1002/hbm.23428

Matthews, P. M., & Jezzard, P. (2004). Functional magnetic resonance imaging. In *J Neurol Neurosurg Psychiatry* (Vol. 75, pp. 6–12).

Melby-Lervåg, M., Redick, T. S., & Hulme, C. (2016). Working Memory Training Does Not Improve Performance on Measures of Intelligence or Other Measures of “Far Transfer”: Evidence From a Meta-Analytic Review. *Perspectives on Psychological Science*, *11*(4), 512–534. https://doi.org/10.1177/1745691616635612

Mercado, E. (2008). Neural and Cognitive Plasticity: From Maps to Minds. *Psychological Bulletin*, *134*(1), 109–137. https://doi.org/10.1037/0033-2909.134.1.109

Minear, M., Brasher, F., Guerrero, C. B., Brasher, M., Moore, A., & Sukeena, J. (2016). A simultaneous examination of two forms of working memory training: Evidence for near transfer only. *Memory and Cognition*, *44*(7), 1014–1037. https://doi.org/10.3758/s13421-016-0616-9

Mitolo, M., Venneri, A., Wilkinson, I. D., & Sharrack, B. (2015). Cognitive rehabilitation in multiple sclerosis: A systematic review. *Journal of the Neurological Sciences*, Vol. 354, pp. 1–9. https://doi.org/10.1016/j.jns.2015.05.004

Morrison, A. B., & Chein, J. M. (2011). Does working memory training work? the promise and challenges of enhancing cognition by training working memory. *Psychonomic Bulletin and Review*, *18*(1), 46–60. https://doi.org/10.3758/s13423-010-0034-0

Nguyen, L., Murphy, K., & Andrews, G. (2019). Cognitive and neural plasticity in old age: A systematic review of evidence from executive functions cognitive training. *Ageing Research Reviews*, Vol. 53, p. 100912. https://doi.org/10.1016/j.arr.2019.100912

Nudo, R. J. (2003). Adaptive plasticity in motor cortex: Implications for rehabilitation after brain injury. *Journal of Rehabilitation Medicine, Supplement*, (41), 7–10.

Nyberg, L., Lövdén, M., Riklund, K., Lindenberger, U., & Bäckman, L. (2012). Memory aging and brain maintenance. *Trends in Cognitive Sciences*, Vol. 16, pp. 292–305. https://doi.org/10.1016/j.tics.2012.04.005

Ophey, A., Roheger, M., Folkerts, A. K., Skoetz, N., & Kalbe, E. (2020). A Systematic Review on Predictors of Working Memory Training Responsiveness in Healthy Older Adults: Methodological Challenges and Future Directions. *Frontiers in Aging Neuroscience*, Vol. 12. https://doi.org/10.3389/fnagi.2020.575804

Pergher, V., Shalchy, M. A., Pahor, A., Van Hulle, M. M., Jaeggi, S. M., & Seitz, A. R. (2020). Divergent Research Methods Limit Understanding of Working Memory Training. *Journal of Cognitive Enhancement*, *4*(1), 100–120. https://doi.org/10.1007/s41465-019-00134-7

Power, J. D., & Schlaggar, B. L. (2017). Neural plasticity across the lifespan. *Wiley Interdisciplinary Reviews: Developmental Biology*, Vol. 6, p. e216. https://doi.org/10.1002/wdev.216

Raymont, V., & Grafman, J. (2006). Chapter 13 Cognitive neural plasticity during learning and recovery from brain damage. *Progress in Brain Research*, Vol. 157, pp. 199–206. https://doi.org/10.1016/S0079-6123(06)57013-X

Redick, T. S. (2019). The Hype Cycle of Working Memory Training. *Current Directions in Psychological Science*, *28*(5), 423–429. https://doi.org/10.1177/0963721419848668

Redick, T. S., Wiemers, E. A., & Engle, R. W. (2019). The role of proactive interference in working memory training and transfer. *Psychological Research*, *1*, 3. https://doi.org/10.1007/s00426-019-01172-8

Reich, D. S., Lucchinetti, C. F., & Calabresi, P. A. (2018). Multiple Sclerosis. *New England Journal of Medicine*, *378*(2), 169–180. https://doi.org/10.1056/NEJMra1401483

Reid, L. B., Boyd, R. N., Cunnington, R., & Rose, S. E. (2016). Interpreting intervention induced neuroplasticity with fMRI: The case for multimodal imaging strategies. *Neural Plasticity*, Vol. 2016. https://doi.org/10.1155/2016/2643491

Rooney, S., Albalawi, H., & Paul, L. (2020). Exercise in the management of multiple sclerosis relapses: current evidence and future perspectives. *Neurodegenerative Disease Management*, *10*(2), 103–115. https://doi.org/10.2217/nmt-2019-0029

Scharfen, J., Jansen, K., & Holling, H. (2018). Retest effects in working memory capacity tests: A meta-analysis. *Psychonomic Bulletin and Review*, *25*(6), 2175–2199. https://doi.org/10.3758/s13423-018-1461-6

Schmicker, M., Schwefel, M., Vellage, A. K., & Müller, N. G. (2016). Training of attentional filtering, but not of memory storage, enhances working memory efficiency by strengthening the neuronal gatekeeper network. *Journal of Cognitive Neuroscience*, *28*(4), 636–642. https://doi.org/10.1162/jocn_a_00922

Schoonheim, M. M., Geurts, J. J. G., & Barkhof, F. (2010). The limits of functional reorganization in multiple sclerosis. *Neurology*, Vol. 74, pp. 1246–1247. Lippincott Williams and Wilkins.

Smitha, K. A., Akhil Raja, K., Arun, K. M., Rajesh, P. G., Thomas, B., Kapilamoorthy, T. R., & Kesavadas, C. (2017). Resting state fMRI: A review on methods in resting state connectivity analysis and resting state networks. *Neuroradiology Journal*, Vol. 30, pp. 305–317. https://doi.org/10.1177/1971400917697342

Solesio-Jofre, E., Beets, I. A. M., Woolley, D. G., Pauwels, L., Chalavi, S., Mantini, D., & Swinnen, S. P. (2018). Age-dependent modulations of resting state connectivity following motor practice. *Frontiers in Aging Neuroscience*, *10*(FEB), 25. https://doi.org/10.3389/fnagi.2018.00025

Soveri, A., Antfolk, J., Karlsson, L., Salo, B., & Laine, M. (2017). Working memory training revisited: A multi-level meta-analysis of n-back training studies. *Psychonomic Bulletin and Review*, *24*(4), 1077–1096.

Tacchino, A., Pedullà, L., Bonzano, L., Vassallo, C., Battaglia, M. A., Mancardi, G., … Brichetto, G. (2015). A New App for At-Home Cognitive Training: Description and Pilot Testing on Patients with Multiple Sclerosis. *JMIR MHealth and UHealth*, *3*(3), e85. https://doi.org/10.2196/mhealth.4269

Teixeira-Santos, A. C., Moreira, C. S., Magalhães, R., Magalhães, C., Pereira, D. R., Leite, J., … Sampaio, A. (2019). Reviewing working memory training gains in healthy older adults: A meta-analytic review of transfer for cognitive outcomes. *Neuroscience and Biobehavioral Reviews*, Vol. 103, pp. 163–177. https://doi.org/10.1016/j.neubiorev.2019.05.009

Thompson, A. J., Baranzini, S. E., Geurts, J., Hemmer, B., & Ciccarelli, O. (2018). Multiple sclerosis. *The Lancet*, Vol. 391, pp. 1622–1636. https://doi.org/10.1016/S0140-6736(18)30481-1

Tomassini, V., Matthews, P. M., Thompson, A. J., Fuglø, D., Geurts, J. J., Johansen-Berg, H., … Palace, J. (2012). Neuroplasticity and functional recovery in multiple sclerosis. *Nature Reviews. Neurology*, *8*(11), 635–646. https://doi.org/10.1038/nrneurol.2012.179

Von Bernhardi, R., Eugenín-Von Bernhardi, L., & Eugenín, J. (2017). What is neural plasticity? In *Advances in Experimental Medicine and Biology* (Vol. 1015, pp. 1–15). https://doi.org/10.1007/978-3-319-62817-2_1

Ward, N. S. (2005). Neural plasticity and recovery of function. *Progress in Brain Research*, Vol. 150, pp. 527–535. https://doi.org/10.1016/S0079-6123(05)50036-0

Willis, S. L., & Schaie, K. W. (2009). Cognitive training and plasticity: Theoretical perspective and methodological consequences. *Restorative Neurology and Neuroscience*, Vol. 27, pp. 375–389. Restor Neurol Neurosci.

World Health Organization. (2020). Rehabilitation. Retrieved March 30, 2021, from https://www.who.int/news-room/fact-sheets/detail/rehabilitation

Zuber, P., Gaetano, L., Griffa, A., Huerbin, M., Bonzano, L., Altermatt, A., … Magon, S. (2021). Additive and interaction effects of working memory and motor sequence training on brain functional connectivity. *Cortex*, (submitted for publication).

Zuber, P., Geiter, E., de Quervain, D., & Magon, S. (2021). Development of a Model-based Working Memory Training and Investigation of its Comparative Efficacy: A Randomized, Double-blind Study on Healthy Adults. *Frontiers in Aging Neuroscience*, (submitted for publication).

Zuber, P., Tsagkas, C., Papadopoulou, A., Gaetano, L., Huerbin, M., Geiter, E., … Magon, S. (2020). Efficacy of inpatient personalized multidisciplinary rehabilitation in multiple sclerosis: behavioural and functional imaging results. *Journal of Neurology*, *267*(6), 1744–1753.