

# Spontaneous neurobiological recovery and modulation of sensorimotor function after ischemic stroke



Sarah Zandvliet





# Spontaneous neurobiological recovery and modulation of sensorimotor function after ischemic stroke

Sarah Zandvliet

The research described in this thesis was supported by two grants of the Dutch brain foundation (Hersenstichting), "EXPLORE-stroke: Exploring plasticity after stroke" (F2011(1)-25) awarded to dr. Meskers, "POTENTIAL: POstural feedback ThERapy combined with Non-invasive TranscranIAL direct current stimulation in patients with stroke" (F2013(1)-41) awarded to dr. van Wegen, and a European advanced grant, 4D-EEG (ERC-AG-ID1, FP7-IDEAS-ERC, 291339) awarded to prof. dr. Kwakkel and prof. dr. van der Helm.

This PhD thesis was embedded within Amsterdam Movement Sciences and Neuroscience research institutes, at the department of rehabilitation medicine, Amsterdam UMC, Vrije Universiteit Amsterdam, the Netherlands.

Financial support by the Dutch Heart Foundation, the Scientific College Physical Therapy (WCF) of the Royal Dutch Society for Physical Therapy (KNGF), the Dutch Institute of Allied Health Care (NPi) and the European Foundation for Health and Exercise (EFOX) for the publication of this thesis is gratefully acknowledged.

Financial support for the publication of this thesis has also been generously provided by TMSi, Motek Medical B.V. and Ipsen Farmaceutica B.V.



Cover	Shutterstock 297171788
Layout	Renate Siebes   Proefschrift.nu
Printed by	ProefschriftMaken
ISBN	978-94-90791-82-7

© 2020 Sarah Zandvliet

All rights reserved. No part of this thesis may be reproduced, stored or transmitted in any form or by any means, without prior permission of the author, or, when applicable, of the publishers of the scientific papers.

VRIJE UNIVERSITEIT

# Spontaneous neurobiological recovery and modulation of sensorimotor function after ischemic stroke

ACADEMISCH PROEFSCHRIFT

ter verkrijging van de graad Doctor  
aan de Vrije Universiteit Amsterdam,  
op gezag van de rector magnificus  
prof.dr. V. Subramaniam,  
in het openbaar te verdedigen  
ten overstaan van de promotiecommissie  
van de Faculteit der Geneeskunde  
op donderdag 29 oktober 2020 om 9.45 uur  
in de aula van de universiteit,  
De Boelelaan 1105

door

**Sarah Bernardina Zandvliet**

geboren te Venray

promotor:

copromotoren:

prof.dr. G. Kwakkel

dr. E.E.H. van Wegen

dr. C.G.M. Meskers

# Contents

Chapter 1	Glossary of terms and general introduction	7
<b>PART 1: SPONTANEOUS NEUROBIOLOGICAL RECOVERY AFTER STROKE</b>		
<hr/>		
Chapter 2	Is recovery of somatosensory impairment conditional for upper limb motor recovery early after stroke?	35
Chapter 3	Is resting-state EEG longitudinally associated with recovery of clinical neurological impairments early post-stroke? A prospective cohort study	61
Chapter 4	Poor motor function is associated with reduced sensory processing after stroke	89
Chapter 5	Position-cortical coherence as a marker of afferent pathway integrity early post-stroke, a prospective cohort study	111
<b>PART 2: MODULATION OF SENSORIMOTOR RECOVERY AFTER STROKE</b>		
<hr/>		
Chapter 6	Short term effects of cerebellar tDCS on standing balance performance in patients with chronic stroke and healthy age matched elderly	147
Chapter 7	The effect of cerebellar transcranial direct current stimulation to improve standing balance performance early post-stroke, study protocol of a randomised controlled trial	177
Chapter 8	General discussion	195
	Summary	215
	Nederlandse samenvatting	227
	Dankwoord (Acknowledgements)	237
	About the author	245
	List of publications	246





# Chapter 1

Glossary of terms and  
general introduction





## GLOSSARY OF TERMS

**Activity:** The execution of a task or action by an individual, see ICF <sup>1</sup>.

**Angiogenesis:** Formation of new blood vessels.

**Bamford classification:** Classification for clinically identifiable subgroups of stroke in order of severity; TACI: total anterior circulation infarcts, PACI: partial anterior circulation infarcts, LACI: lacunar infarcts <sup>2</sup>.

**Behavioural restitution of function:** "The return towards more normal patterns of motor control with the impaired effector (a body part such as the hand or foot that interacts with an object or the environment) and reflects the process towards true (neurological) recovery" <sup>3</sup>. Behavioural restitution of function and **behavioural substitution/compensation of function** are contrary mechanisms that can both contribute to improvement on an **activity** level.

**Behavioural substitution/compensation of function:** Completing a task using alternative effectors, joints, muscles or kinematics. That is to say in a manner that is qualitatively different from healthy controls <sup>4</sup>.

**(Bio)marker:** A characteristic that is objectively measured and evaluated as an indicator of normal biological processes, pathogenic processes, or responses to a therapeutic intervention <sup>5</sup>.

**Body function:** A physiological function of a body system, including psychological functions, see ICF <sup>1</sup>.

**Connectivity:** Connected structures or connective activity patterns in the brain. Structural connectivity refers to brain regions connected by anatomical structures. Functional connectivity refers to related activity of brain regions <sup>6</sup>.

**Coherence:** Resemblance between two signals, in this thesis it is used in the context of **system identification** <sup>7</sup>.

**Cortical reorganisation:** A stroke will lead to changes in neuronal activity, connectivity structure and the bodies representation in the cortex (cortical map) due to the damaged tissue, loss of limb function and repair mechanisms <sup>8</sup>.

**Diaschisis:** Distant neurophysiological and metabolic changes caused by a focal injury <sup>9,10</sup>.

**Electroencephalography (EEG):** Is used to record the electrical activity of neurons in the brain. It is measured by placing electrodes onto the skull. EEG can also be measured invasively, in which the electrodes are placed directly on the brain tissue, this is referred to as intracranial EEG (iEEG) <sup>11</sup>.

**(End) effector:** A body part such as the hand or foot that interacts with an object or the environment <sup>4</sup>.

**Experience/learning-dependent plasticity:** Synaptic plasticity due to changes in environment or behaviour see also **neural plasticity** <sup>12,13</sup>.

**Hebbian learning:** Theory formed by Hebb in 1949, which is currently the most important neurophysiological learning theory based on synaptic plasticity, see **neural plasticity**. It states that neurons that will consistently fire together, will lead to an increase in efficiency which is needed for metabolic changes and growth processes <sup>14</sup>. Anti-Hebbian learning refers to a loss of efficiency due to an opposite process. Non-Hebbian learning refers to forms of neural plasticity that cannot be explained by the Hebbian learning theory.

**International classification of functioning, disability, and health (ICF):** A classification that provides a standard language and conceptual basis for the definition and measurement of health and disability <sup>1</sup>.

**Ischemic stroke:** An episode of neurological dysfunction caused by focal cerebral, spinal, or retinal infarction. The infarction is caused by arterial thrombotic blockage leading oxygen deprivation in the central nerve system <sup>15</sup>. See **stroke** for the definition of a central nerve system infarction.

**Long term depression (LTD):** Decreasing activity leading to long lasting changes in synaptic connections. Since this process is established in animal models, this is referred to as LTD-like plasticity in humans. See **neural plasticity** <sup>16</sup>.

**Long term potentiation (LTP):** Increasing activity leading to long lasting changes in synaptic connections. Since this process is established in animal models, this is referred to as LTP-like plasticity in humans. See **neural plasticity** <sup>16</sup>.

**Magnetic resonance imaging (MRI):** Medical imaging technique using a magnetic field to generate images of organs. Functional MRI (fMRI) makes use of the blood-oxygen-level-dependent (BOLD) signal to observe activity related decreases in oxygenation of blood in brain areas of interest <sup>17</sup>.

**Magnetoencephalography (MEG):** Is a technique for recording brain activity by recording the magnetic fields of the electrical activity of the brain <sup>18</sup>.

**Motor control:** Motor control is the process by which motor commands produced by the central nerve system activate and coordinate muscles to generate joint torques to move effectors in goal-directed actions <sup>19</sup>.



**Motor function:** The physiological **body function** to produce force, move a body part, or maintain a posture under external disturbance, see ICF <sup>1</sup>.

**Motor recovery:** Improvement in motor performance dependent on the tasks and measures that are used <sup>20</sup>.

**Neural networks:** Cortical and subcortical areas of neuronal populations that interact with each other <sup>21</sup>.

**Neural plasticity:** The ability of the brain to change and adapt continuously throughout the human lifespan. These changes occur to optimize the neuronal network, thereby strengthen or weaken specific functions. Synaptic plasticity is the strengthening or weakening of synaptic connections between neurons, whereas non-synaptic plasticity refers to the excitability changes within the neuron <sup>12,13,16,22</sup>.

**Neuronal (brain) oscillations:** Are rhythmic or repetitive patterns of neural activity in the brain. Neural tissue can generate oscillatory activity, driven by mechanisms within individual neurons or by interactions between neurons. The synchronized activity of large numbers of neurons can give rise to oscillations, which can be observed in an **electroencephalogram (EEG)** <sup>23</sup>.

**Non-invasive brain stimulation:** Techniques used for modulation of the nervous system. Examples are: **transcranial magnetic stimulation (TMS)**, **transcranial direct current stimulation (tDCS)**, **transcranial electric stimulation (tES)**, and **transcranial alternating current stimulation (tACS)**.

**Paralysis:** Inability to move part of the body.

**Paresis:** Weakness of a part of the body.

**Participation:** Involvement in a life situation, see ICF <sup>1</sup>.

**Penumbra:** Damaged tissue due to hypoxia following a stroke in danger of necrosis, yet still reversible when oxygen supply can be restored <sup>24</sup>.

**Phases in stroke recovery:** Hyper-acute: 0–24 hours, acute: 1–7 days, early subacute: 7 days–3 months, late subacute: 3–6 months, chronic: >6 months <sup>3</sup>.

**Posture:** The physiological body function to remain in a static vertical position, i.e. standing and sitting.

**Quality of movement:** Is the direct comparison of a stroke patient's motor execution to healthy age-matched controls. The closer one approaches normal motor control the higher the quality of the movement <sup>25</sup>.

**Reactive/non-learning-dependent plasticity:** A stroke triggers a cascade of hemodynamic and neuroinflammatory reactions, responsible for true neurological repair within the brain. These reactive mechanisms included recovery of the **penumbral tissue**, **decreasing diaschisis**, **angiogenesis**, homeostatic mechanisms including increased secretion of **growth factors and neurotransmitters** that lead to enhanced gene expression, axonal sprouting of remaining neurons and increased synaptic activity<sup>12,26</sup>. These reactive mechanisms, leading to **neural plasticity**, are referred to as reactive plasticity within this thesis.

**Recoverer/non-recoverer:** Classification to distinguish the longitudinal recovery pattern of a patient between the acute and chronic phase after stroke. The terms **fitter/ non-fitter** refer to the same concept<sup>27</sup>.

**Sensorimotor function:** Central and peripheral integration of sensory (**somatosensory**, auditory and visual) and **motor function** needed for optimal execution of a movement with a particular **effector** in a specific task context.

**Somatosensory impairment:** A loss in the body function to sense: touch, temperature, pain, position and movement (proprioception), and recognition of an object through touch<sup>28</sup>. See ICF<sup>1</sup>.

**Spectral characteristics:** Quantification of the strength of the neuronal oscillations in terms of spectral power, measured with EEG or MEG, in a specific frequency band or ratio.

**Spontaneous (neuro)biological recovery:** Improvements in recovery of behaviour, occurring during a time-sensitive window of heightened recovery that begins early after stroke and slowly tapers off<sup>3</sup>.

**Standing balance:** The physiological body function to remain in a static standing position, within this thesis, the term refers to bilateral standing.

**Stroke:** The broad term stroke refers to a central nerve system (CNS) infarction in the brain, spinal cord or retinal cell death attributable to ischemia. The diagnose is based on: Pathological, imaging, or other objective evidence of cerebral, spinal cord, or retinal focal ischemic injury in a defined vascular distribution; or clinical evidence of cerebral, spinal cord, or retinal focal ischemic injury based on symptoms persisting  $\geq 24$  hours or until death, when other etiologies are excluded<sup>15</sup>.

**System identification:** Mathematical method to quantify a system originating from the engineering field. By measuring the output from a system, during or after a known input, the interaction between the input and the motor control system can be quantified. The given input is also referred to as perturbation<sup>7</sup>.

**Task performance:** Execution of a given task, within this thesis it generally refers to a motor task.

**Transcranial direct current stimulation (tDCS):** Is a form of non-invasive brain stimulation in which a constant, low direct current flows via electrodes into the skull. In contrast to **TMS**, tDCS does not lead to the generation of an action potential. The electrode(s) from which the current flows and therefore positively charged is the anode, the electrode(s) towards which the current flows is negatively charged and called the cathode <sup>29</sup>.

**Transcranial magnetic stimulation (TMS):** Is a form of non-invasive brain stimulation in which electromagnetic induction is used to cause an electric current to flow in a small targeted region of the brain. A magnetic field generator called a coil, is placed on the scalp which delivers a changing electric current to the coil. This pulse is strong enough to generate an action potential in the motor cortex leading to a contraction, of, for example, a hand muscle <sup>30</sup>.

## GENERAL INTRODUCTION

### Stroke

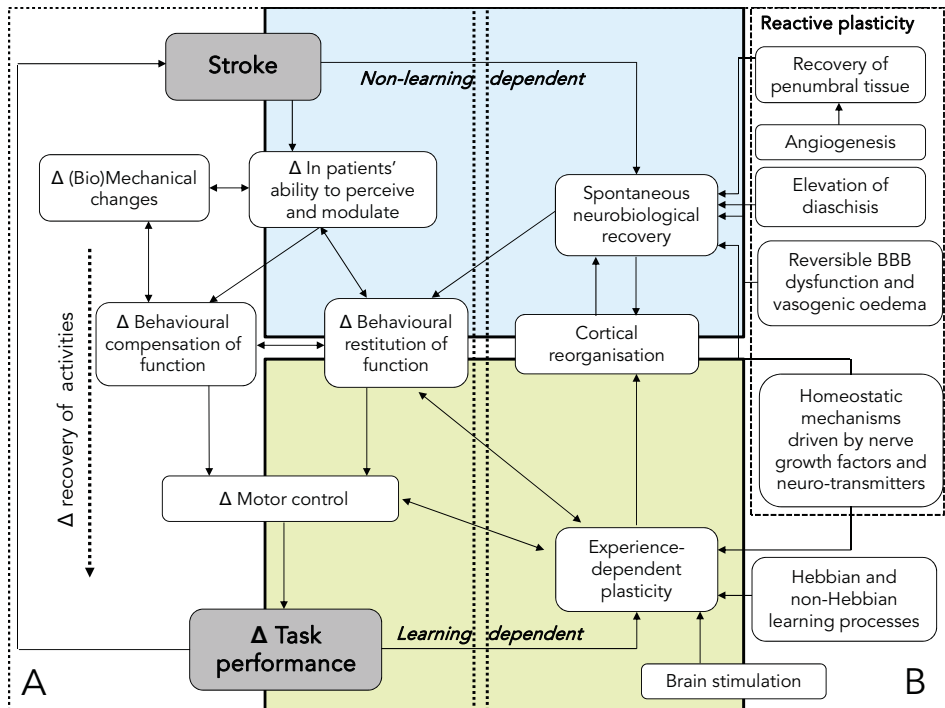
An ischemic stroke is a central nervous system infarction due to arterial thrombotic blockage<sup>15</sup>. The diagnose is based on: pathological, imaging, or other objective evidence of ischemic injury or clinical evidence based on overt symptoms like paresis of the arm and leg, and the inability to comprehend and produce speech that remain present more than 24 hours after onset<sup>15</sup>.

This thesis focusses on cerebral ischemic stroke, as a common subtype of stroke, which accounts for 87% of stroke incidents<sup>15,31</sup>. Within a specific time window after artery blockage, thrombolysis with recombinant tissue plasminogen activator or thrombectomy can be applied to restore oxygenated regional cerebral blood flow and prevent cell death<sup>32,33</sup>. These curative interventions can thereby prevent death and severe disability of patients. "Time is brain", acute care is therefore focussed on reducing the time from stroke to initiation of above mentioned curative interventions<sup>34</sup>. Currently, these curative interventions are still limited to only those patients who had no previous stroke in the last 3 months, having no haemorrhagic signs and who are medically stable enough to receive treatment within a maximum of 6–7 hours after onset<sup>33,35</sup>. Despite these developments in restorative intervention, it is estimated that thrombolysis is only effective in 1 out of 5 patients<sup>36</sup>, whereas 1 out of 8 patients benefits from endovascular treatment<sup>37</sup> following the modified ranking scale. Above findings suggest that most patients remain affected in terms of body function, such as speech, motor, somatosensory and cognitive functions, as well as on an activity and participation level, such as motor activities, mobility, self-care and communication<sup>1</sup>. Task-specific and individualized rehabilitation strategies tailored to optimize meaningful activities in daily living such as standing, balance, gait, and upper limb activities are therefore needed<sup>38</sup>. This thesis aimed to increase knowledge on how underlying neuronal mechanisms relate to processes that contribute to sensorimotor recovery after stroke as displayed in Figure 1.1.

### Processes underlying sensorimotor recovery after stroke

Figure 1.1 panel A, shows a phenomenological model of the assumed process in the recovery of activities after stroke<sup>26,40</sup>. Prospective cohort studies in the field of stroke recovery so far show that a majority of patients with a first ever hemispheric stroke show some degree of recovery of the affected modalities<sup>41</sup>. Unilateral loss of motor function of the upper limb affects many aspects of daily living and is one of the most common impairments affecting about 75% of all stroke survivors<sup>42</sup>. By intensive motor training, one can (re)learn





**Figure 1.1** | Phenomenological model adapted from Kwakkel et al. 2004 and Buma et al. 2013<sup>26,39,40</sup>. Panel A shows the assumed processes underlying recovery of activities after stroke. Panel B shows the assumed underlying neuronal mechanisms in recovery of activities after stroke. The lines represent a relationship between two mechanisms and do not necessarily refer to a causal relationship.  $\Delta$  symbol represents change, BBB: blood-brain barrier. This thesis aimed to increase knowledge on how underlying neuronal mechanisms, in panel B, relate to the processes that contribute to recovery of activities after stroke, as displayed in panel A, focusing on sensorimotor impairments. The first part of this thesis, blue panel, focusses on non-learning-dependent mechanisms and spontaneous neurobiological recovery after stroke. The second part of this thesis, green panel, focusses on learning-dependent mechanisms and the potential for modulation of sensorimotor recovery after stroke by brain stimulation.

to compensate for normal motor function with alternative effectors to execute a task, i.e. behavioural compensation of function, yet with little generalisation to other tasks<sup>4,38</sup>. Motor learning is required for improvement in performance of meaningful activities such as grasping and walking. However, these improvements are dependent on a patient's ability to perceive, effecting the ability to modulate an end-effector and with that, may be compromised by somatosensory impairments<sup>26,43</sup>. Incomplete behavioural restitution of sensorimotor impairments will force patients to compensate by using their end-effectors in a different way to accomplish daily tasks. There is growing evidence that a return of quality of movement as a reflection of behavioural restitution, quantified by kinematic

measures<sup>44-47</sup>, is driven by poorly understood mechanisms of spontaneous neurobiological recovery within the first 10 weeks after stroke onset<sup>43,48</sup>. However, a patient's ability to control movement is not only determined by neuronal processes but also by gradual biomechanical changes in the movement apparatus. The impact of these biomechanical changes such as enhanced stiffness and muscle length, change the perception and modulation of end-effectors during performance of meaningful activities<sup>49-51</sup>, Figure 1.1, panel A.

## Neuronal mechanisms underlying recovery after stroke

### *Non-learning-dependent mechanisms*

Based on animal studies, there is strong evidence that a cascade of hemodynamic and neuroinflammatory reactions are responsible for true neurological repair in the brain that starts as a reaction to the infarction within minutes to hours after stroke onset<sup>12,26,27</sup>. A schematic overview of these mechanisms, proposed as a model for recovery of activities after stroke is displayed in Figure 1.1, panel B<sup>26,40</sup>. The main endogenous mechanisms of repair include salvation of penumbral tissue<sup>24,52</sup> and alleviation of connected suppressed areas remote from the infarcted area, i.e. alleviation of diaschisis<sup>9,10</sup>. These metabolic processes of neuronal recovery are believed to be enhanced by an increased gene-expression of growth promoting factors such as brain derived neurotrophic factor (BDNF)<sup>53,54</sup>. As a consequence, the enhanced synapto(neo)genesis of survived and remaining neurons may contribute to repair and adaptive neuronal networks in the brain<sup>12,55-57</sup>. These reactive mechanisms triggered by the stroke, leading to neural plasticity, are collectively referred to as reactive plasticity within this thesis. After 2 to 4 weeks, animal studies show an increased gene-expression of growth inhibiting factors, such as NOGO-A, closing the time window with an increased potential for spontaneous neurobiological recovery<sup>12</sup>. Due to the optimal conditions of enhanced neural plasticity in this specific early time window after stroke, it is also considered as the optimal time window for intervention to promote enhanced recovery. There is evidence that the upregulation of growth inhibiting factors is postponed by upper limb training in animals<sup>58</sup>. However it is not known if and by which type of intervention neural plasticity can be influenced in humans<sup>59</sup>.

### *Learning-dependent mechanisms*

Besides reactive plasticity induced by the stroke, neuronal changes can also occur due to changes in environment or behaviour, for example in learning a motor task. These neuronal changes in the brain occur throughout the whole human lifespan and can strengthen or weaken specific functions that are referred to as experience-dependent plasticity.

Experience-dependent plasticity is theorized to be the major underlying neurophysiological mechanism of learning, called the Hebbian theory<sup>14</sup>. Neuron 'A' needs to consistently take part in firing neuron 'B' in order to accomplish growth processes or metabolic changes such that A's efficiency, as one of the cells firing B, is increased<sup>14</sup>. Or paraphrased more simply: 'Neurons that fire together, wire together'<sup>60</sup>. This process is mediated by strengthening or weakening of synaptic connections through alternation in the synaptic activity of neuronal networks<sup>12,13,16,22</sup>. When process leads to long-lasting changes in synaptic connections, this is referred to as long-term potentiation (LTP) and long-term depression (LTD) in animals and LTP-like and LTD-like plasticity in humans<sup>16</sup>. Hebbian learning cannot explain all forms of neural plasticity. Improvement in motor performance and for example intra limb coordination can also be explained by non-Hebbian forms of neural plasticity due to an enhanced collateral, surrounding inhibition and with that improving the precision in modulation, Figure 1.1<sup>16,22</sup>. Sustained input that is not important can, for example, lead to shrinkage of the cortical representation of whiskers in rodents, as a mechanism to normalize firing rate<sup>22,61</sup>. An enriched environment also causes a shrinking/focussing in the cortical whisker representation in rodents, which is attributed to the upregulation of arousal related modulators and can also not be explained by Hebbian forms of plasticity<sup>22,62</sup>.

## **PART 1: Spontaneous neurobiological recovery after stroke**

Due to the above described mechanisms of reactive plasticity in the brain, stroke patients can show rapid neurological recovery<sup>12</sup>. While the majority of patients show at least some level of motor recovery of the upper limb, about 30% of patients show no to very little improvement<sup>63</sup>. For this specific group, the so called non-recoverers, an accurate individual prediction of a patient's potential for recovery is unfortunately not yet possible<sup>64</sup>. This step towards the individual prediction of recovery for all patients is needed for clinical care, as well as for patient selection in clinical trials to develop the optimal treatment for different subsets of patients<sup>65</sup>. The first part of this thesis focusses on non-learning-dependent mechanisms and aims to find potential clinical and neurophysiological markers for spontaneous neurobiological recovery in patients with sensorimotor impairments after an ischemic stroke, Figure 1.1, blue panel.

### **Clinical markers of spontaneous neurobiological recovery**

Nijland et al. found that patients who are able to produce some shoulder abduction and finger extension within 72 hours post-stroke have a 98% chance of regaining some dexterity, defined as a minimal of 10 out of 57 points on the action research arm test

(ARAT) <sup>66</sup>. This model is known as the SAFE model <sup>67</sup>. Patients who are unable to produce these movements within 72 hours still have a 25% chance of regaining some dexterity. The probability for a patient to regain some function decreases over time and is very small after 12 weeks post-stroke <sup>68</sup>.

### *Proportional recovery model*

Prabhakaran et al. first described spontaneous neurobiological recovery in 41 patients with a stroke as being proportional to the initial impairment <sup>69</sup>. They found in the majority of patients a proportional recovery of approximately 70% of their potential measured on the Fugl-Meyer motor assessment of the upper extremity (FM-UE), i.e.  $\text{potential recovery} = 0.7 * (\text{maximal FM-UE score (66 points)} - \text{initial measurement score}) + 0.4$ . This linear regression model aimed to explain the mathematical relationship between initial FM-UE and improvement was confirmed by Winters et al. <sup>63</sup> in a larger subset of 211 patients with a stroke and applied to several other domains including the lower limb <sup>70,71</sup>, aphasia <sup>72</sup> and visual spatial neglect <sup>73,74</sup>. Most of these studies identified a subpopulation of stroke patients who failed to show the predicted proportional recovery. It is not known why this subset of patients fails to show spontaneous neurobiological recovery. Yet, patients who fail to show recovery of the upper limb, so-called non-recovers or non-fitters, more often have an affected leg function, a facial paralysis, no finger extension, larger subcortical lesions, classification as more severely affected (Bamford classification) and are older than patients who do show spontaneous neurobiological recovery <sup>63,69</sup>. In order to set realistic treatment goals and provide patients with optimal care, it is important to identify this group early post-stroke.

### *Somatosensory impairments*

The ability to process sensory information, like touch, pain and body position, is affected in between 34–84% of stroke survivors <sup>75–78</sup>. Even though somatosensory impairment is related to impaired motor outcome, and is likely to be essential for motor recovery, it has to date received very limited attention <sup>75,76,79</sup>. Prospective cohort studies aimed to assess somatosensory impairments as well as its time course of recovery are lacking. It is unknown whether post-stroke upper limb motor and somatosensory impairment reflect a parallel process of spontaneous neurobiological recovery in both modalities or if somatosensory impairment and/or recovery influences behavioural restitution of motor function. In **chapter 2** of this thesis, the association between motor and somatosensory impairments was longitudinally investigated.



## Neurophysiological markers of cortical reorganisation

For the subgroup of severely affected patients, who are unable to produce any voluntary movement of the affected limb, it is especially important to find markers of cortical reorganisation since clinical scales fail to provide accurate prediction of outcome. Markers directly derived from brain measures could possibly give insight in cortical reorganisation occurring in the brain<sup>80</sup>. These markers might serve as predictors of spontaneous neurobiological recovery, days or weeks before a patient is showing behavioural restitution of function of the upper limb, and could help to improve early prediction of outcome.

The integrity of the cortical spinal tract (CST) is considered a prerequisite for good motor function of the upper limb. If the pathway is intact, an action potential will be conducted along the CST and the descending nerve, causing a contraction of the muscle<sup>81,82</sup>. This intactness can be measured with an assessment of a motor evoked potential (MEP) induced by transcranial magnetic stimulation (TMS) and could be a potential marker in post-stroke prediction models<sup>30</sup>. Even though this method does not perform better in predicting long term motor recovery compared to clinical assessment on a group level, it can be a beneficial marker for prediction of outcome specifically in patients with severe motor impairments in the first weeks post-stroke<sup>80,83-86</sup>.

### *Magnetic resonance imaging*

Magnetic resonance imaging (MRI) is used to longitudinally investigate changes in brain networks, which could lead to a potential marker for prediction of recovery<sup>87</sup>. Changes in specific network connectivity have been associated with spontaneous recovery of language, attention, motor and somatosensory impairments<sup>88-94</sup>. Nijboer et al. however found no relation between network connectivity during an awake state of rest and upper limb motor recovery in a longitudinal study in 13 stroke patients<sup>87</sup>.

### *Electroencephalography and magnetoencephalography*

Electroencephalography (EEG) and magnetoencephalography (MEG) can be used to measure cortical activity with a high temporal resolution during a specific task in the order of milliseconds. EEG provides a suboptimal spatial resolution due to volume conduction of the skull as compared to MEG. EEG can, however, be measured during a balance task and larger movements of the arm and hand which is not possible with MEG.

Neuronal oscillations of specific frequencies, measured with EEG or MEG, have been related to specific cortical functions<sup>23</sup>. Alpha waves, neuronal oscillations in the 8–12 Hz frequency band, are suggested to reflect a state of increased learning and optimal concentration<sup>95</sup>. Neuronal oscillations can also disclose pathologies: a decrease in alpha

activity in rest is generally seen in patients after stroke with a co-occurring increase in spectral power of slow frequency oscillations like theta (4–8 Hz) and delta (1–4 Hz)<sup>23,96,97</sup>. The strength of the neuronal oscillations quantified with spectral characteristics has also been investigated after stroke. Asymmetry in spectral characteristics between the hemispheres has been associated with poor clinical outcome at 6 months post-stroke and is believed to reflect the severity of the neurological damage shortly after stroke<sup>98,99</sup>. Spectral characteristics derived from EEG during rest can potentially be used as a representation of the severity of the damage and cortical reorganisation, which influences the amount of spontaneous neurobiological recovery taking place after the stroke. In **chapter 3** the longitudinal changes in EEG based spectral characteristics measured during post-stroke recovery, were investigated. It was hypothesised that an increase in slow oscillations and a relatively larger asymmetry in spectral power between hemispheres in the first weeks after stroke were related to the stroke severity and motor function of the upper limb.

To specifically study the cortical involvement in movement execution in patients with a stroke, an approach is required that is closely related to the execution of a motor task, yet is also feasible in patients without voluntary motor control of the limb. System identification techniques can be used to study the cortical involvement in motor control<sup>7</sup>. Perturbations applied to the affected arm can provide information on the processing of the signal within the motor cortex, without requiring voluntary movement of the patient. The coherence between a position perturbation and the cortical response is therefore a potential neurophysiological marker related to spontaneous neurobiological recovery. In **chapter 4 and 5** of this thesis, position-cortical coherence was examined and hypothesized as a potential neurophysiological marker of somatosensory pathway integrity and spontaneous neurobiological recovery.

## **PART 2: Modulation of sensorimotor recovery after stroke**

The second part of this thesis focusses on learning-dependent mechanisms and aimed to investigate the potential to modulate sensorimotor recovery with non-invasive brain stimulation, Figure 1.1, green panel.

### **Transcranial direct current stimulation**

Electrical stimulation on the skull has been applied for centuries for the treatment of a wide variety of diseases. The method was reintroduced about 20 years ago by Nitsche and Paulus to increase neuronal excitability in the brain in the area of stimulation and has

since then found many applications<sup>29,100</sup>. In transcranial direct current stimulation (tDCS) a weak direct current (up to 3 mA) flows through electrodes placed onto the skull. In contrast to TMS, tDCS cannot directly elicit an action potential<sup>100</sup>. tDCS is assumed to induce polarity specific alterations in the membrane potential of neurons which will enhance or reduce the calcium influx via N-methyl-D-aspartate (NMDA) receptors and voltage-gated calcium channels<sup>100-102</sup>. The direction of synaptic changes is dependent on the amount of intracellular calcium, in which cathodal stimulation will generally induce low and anodal stimulation will generally induce high concentration levels, leading to respectively decreased or increased excitability changes and LTD- or LTP-like plasticity<sup>100,103</sup>. When stimulation is not leading to a distinct high or low concentration, or in case of a calcium overflow, no clear direction in synaptic changes occurs<sup>104-106</sup>. These effects of tDCS seem to be modulated by gamma-Aminobutyric acid (GABA)<sup>107-109</sup>. Other neurotransmitters and modulators such as dopamine and acetylcholine have been found to have an effect on tDCS induced neural plasticity<sup>110</sup>, however, the exact mechanisms are not yet understood. tDCS is also found to upregulate the secretion of brain derived neurotrophic factor (BDNF)<sup>111,112</sup>, which could further enhance synaptic plasticity<sup>113</sup>. Specifically, how BDNF is enhanced in the above described cascade needs further investigation.

### *tDCS after stroke*

tDCS has been extensively investigated as a tool to optimize rehabilitation after stroke, in which the majority of studies have focused on the upper limb<sup>114,115</sup>. Current meta-analyses indicate a low to moderate quality of evidence for improving ADL performance with tDCS applied to the motor cortex after stroke and no evidence for the effectiveness of tDCS to enhance recovery of the upper and lower limb<sup>116</sup>. The meta-analysis performed by Marquez et al. indicated that while no added value of tDCS was found at the group level, small significant improvements were found when stroke characteristics were taken into account<sup>117</sup>. These results indicate that tDCS research needs to focus on interindividual differences to direct research towards those patients in which tDCS could potentially have an added value<sup>115</sup>. Which interindividual characteristics and related underlying mechanisms are important is not established.

### *Cerebellar tDCS*

The cerebellum plays a distinct role in motor adaptation through error-based learning and has often been described as a controller of temporal and spatial movement accuracy<sup>118-121</sup>. Around 80% of all neurons in our brain are located in the cerebellum<sup>122</sup>. Both these facts may serve as a basis for testing the therapeutic impact of tDCS on the large pool of Purkinje cells within the cerebellum, to enhance motor adaptation. To apply cerebellar (cb)-tDCS,

one electrode is placed on the cerebellar hemisphere and the second electrode is placed on the cheek where it has no influence on neurons<sup>123</sup>. This setup has an optimal current flow to target the underlying cerebellar areas as compared to typically used cortical setups<sup>123</sup>. Anodal stimulation on the cerebellar hemisphere leads to increased cerebellar brain inhibition (CBI) at the contralateral cortical hemisphere and has been related to synaptic based forms of learning<sup>118,124–126</sup>.

The interplay between the motor cortex and cerebellum is vital for both skill learning and motor adaptation, yet not fully understood<sup>127</sup>. It is evident that the cerebellum has an important function in timing, in which a strong motor cortex to cerebellar connection leads to more accurate movement endpoints<sup>127,128</sup>. The cerebellar hemispheres contain a large amount of Purkinje cells which play an important role in feedback-based learning. Parallel and climbing fibers in the cerebellum mediate the process of enhanced activation of Purkinje cells and modulate motor execution via the cerebello-thalamo-cortical pathway<sup>129,130</sup>. The cerebellum and cerebrum are connected via the cerebello-thalamo-cortical pathway, which propagates efferent signals from the cerebellum through the contralateral thalamus to the cerebrum. Afferent, sensory information is arriving at the cerebellum via the cortico-ponto-cerebellar pathway<sup>121,131</sup>. Anodal cb-tDCS is thought to enlarge the population of activated Purkinje cells, leading to a larger involvement of the cerebellum in the executed motor task<sup>132</sup>. The complex interplay of LTD and LTP of Purkinje cells in the cerebellum could lead to an increase of synaptic based plasticity<sup>16,118,125,126</sup>.

### ***Mechanisms of action of cerebellar tDCS in patients with a stroke***

Recently, there has been increased attention for the possible beneficial effects of cb-tDCS in post-stroke motor function recovery as an alternative for cortical stimulation. When cortical areas are affected by a stroke, it might be more beneficial to optimize the undamaged cerebellum, than to target the lesioned area directly where it is unclear which structural and functional pathways are intact or not<sup>133</sup>. The mechanism of upregulation of BDNF in the cerebellum could lead to enhanced motor learning after stroke<sup>111,119,130,134</sup>.

### ***Cerebellar tDCS to improve balance performance***

Impaired standing balance after stroke is common and causes fall events, hampers walking capacity and increases ADL dependence, and is therefore an important target for rehabilitation interventions<sup>135</sup>. The cerebellum has an important function in balance control, yet since this function is located more in the midline and anterior parts of the cerebellum it can likely not be targeted directly with tDCS<sup>121,123,136,137</sup>. However, improvements in standing balance performance can potentially be achieved by enhancing cerebellar

function in motor adaptation and stimulate experience-dependent plasticity during standing balance training. Since anodal tDCS is only enhancing excitability, it needs to be applied in combination with a challenging task to enhance motor learning<sup>138</sup>. To accurately measure improvements in standing balance performance kinetic or kinematic measures are needed, which are responsive for change and can capture the quality of movement control<sup>25</sup>. **Chapter 6** examined the effects of two forms of cerebellar tDCS for enhancing motor adaptation and therefore balance performance in patients with a chronic stroke and a healthy control group.

## **Modulation of sensorimotor recovery in the time window of spontaneous neurobiological recovery by cerebellar tDCS**

### *Investigating the interaction of learning and non-learning-dependent mechanisms*

Experience-dependent plasticity influences cortical representations of function in the undamaged tissue and might be able to directly enhance spontaneous neurobiological recovery<sup>139</sup>. A series of roundtable expert meetings (stroke recovery and rehabilitation roundtable) has indicated the search for interaction effects between therapy and spontaneous neurobiological recovery as a prime target for new research<sup>25,43,80,140,141</sup>.

The time window in the early phase after stroke in which reactive plasticity occurs may provide a unique time period for the enhancement of BDNF-mediated, LTP-like plasticity by application of cerebellar tDCS. Only clinical trials in the early phase post-stroke can determine if enhanced experience-dependent plasticity by cerebellar tDCS can induce an interaction effect with spontaneous neurobiological recovery, enhancing behavioural restitution of function. In **chapter 7**, the protocol of a randomized controlled trial to study the effect of cerebellar tDCS in the time window of spontaneous neurobiological recovery is described.

## **Contributions to the phenomenological model**

This thesis aimed to strengthen the phenomenological model presented in Figure 1.1 by several studies that investigated the connections between underlying neuronal mechanism, panel 1B, to processes that contribute to recovery of activities after stroke, panel 1A, focusing on sensorimotor impairments. **The first part of this thesis, Figure 1.1 blue panel**, focusses on non-learning-dependent mechanisms and spontaneous neurobiological recovery after stroke. In **chapter 2**, the influence of a patients' ability to perceive and modulate, in terms of somatosensory impairments, on behavioural restitution of upper limb motor function was investigate. It was hypothesized that improvements in

both motor and somatosensory impairments were largely driven by a common process of spontaneous neurobiological recovery. **Chapter 3–5** investigated different possible markers of cortical reorganisation from neurophysiological recording, (EEG), before they translate into behavioural restitution of motor function of the upper limb. In **chapter 3** the longitudinal changes in EEG based spectral characteristics measured during post-stroke recovery, were investigated. It was hypothesized that an increase in slow oscillations and a relative larger asymmetry in spectral power between hemispheres in the first weeks after stroke were related to the stroke severity and motor impairment of the upper limb. In **chapter 4 and 5** of this thesis, the coherence between a position perturbation and the cortical response (PCC) was examined. The presence of PCC was hypothesized to be a potential neurophysiological marker of somatosensory pathway integrity and spontaneous neurobiological recovery.

The second part of this thesis, **Figure 1.1 green panel**, focusses on learning-dependent mechanisms and the potential for modulation of sensorimotor recovery after stroke by brain stimulation (cb-tDCS). **Chapter 6** focused on the effect of brain stimulation on experience-dependent plasticity. To investigate if via this pathway task performance could be improved in chronic stroke patients in terms of standing balance performance. In **chapter 7**, the protocol of a randomized controlled trial to study the effect of cerebellar tDCS in the time window of spontaneous neurobiological recovery is described. This study is designed to investigate a possible influence of experience-dependent plasticity on behavioural restitution of function. **Chapter 8** concludes with an overview and general discussion of the found results and recommendations for future research.

The research in this thesis was financed by the grants, “EXPLORE-stroke: Exploring plasticity after stroke” (F2011(1)-25) awarded to dr. Meskers, “POTENTIAL: POstural feedback ThErapy combined with Non-invasive TranscranIAL direct current stimulation in patients with stroke” (F2013(1)-41) awarded to dr. van Wegen, by the Dutch brain foundation (Hersenstichting), and a European advanced grant, 4D-EEG (ERC-AG-ID1, FP7-IDEAS-ERC, 291339) awarded to prof. dr. Kwakkel and prof. dr. van der Helm.

The chapters of this thesis are based on the data collected within the EXPLORE-stroke, 4D-EEG and POTENTIAL projects and the “EXPLICIT-stroke: EXplaining PLasticity after stroke” trial funded by the Dutch organisation for health research and development (ZonMw, grant number 89000001) awarded to prof. dr. Kwakkel.



## REFERENCES

- 1 World Health Organization. International Classification of Functioning, Disability and Health. Geneva, 2001.
- 2 Bamford J, Sandercock P, Dennis M, Warlow C, Burn J. Classification and natural history of clinically identifiable subtypes of cerebral infarction. *Lancet* 1991; **337**: 1521–1526.
- 3 Bernhard J, Kwakkel G, Lannin NA, Borschmann K, English C, Ali M et al. Consensus Statements from the Stroke Recovery and Rehabilitation Roundtable standardized measurement of sensorimotor recovery in stroke trials: Consensus-based core recommendations from the Stroke Recovery and Rehabilitation Roundtable. *J Stroke* 2017; **31**: 784–792.
- 4 Levin MF, Kleim JA, Wolf SL. What do motor ‘recovery’ and ‘compensation’ mean in patients following stroke? *Neurorehabil Neural Repair* 2009; **23**: 313–319.
- 5 Biomarkers Definitions Working Group. Biomarkers and surrogate endpoints: Preferred definitions and conceptual framework. *Clin Pharmacol Ther* 2001; **69**: 89–95.
- 6 Grefkes C, Fink GR. Connectivity-based approaches in stroke and recovery of function. *Lancet Neurol* 2014; **13**: 206–216.
- 7 Meskers CGM, de Groot JH, de Vlugt E, Schouten AC. NeuroControl of movement: system identification approach for clinical benefit. *Front Integr Neurosci* 2015; **9**: 48.
- 8 Wittenberg GF. Experience, cortical remapping, and recovery in brain disease. *Neurobiol Dis* 2010; **37**: 252–258.
- 9 Carrera E, Tononi G. Diaschisis: past, present, future. *Brain* 2014; **137**: 2408–2422.
- 10 Feeney D, Baron J. Diaschisis. *Stroke* 1986; **17**: 817–830.
- 11 Niedermeyer E, Lopes da Silva FH. *Electroencephalography: basic principles, clinical applications, and related fields*. 7th editio. Oxford University Press, 2005.
- 12 Murphy TH, Corbett D. Plasticity during stroke recovery: from synapse to behaviour. *Nat Rev Neurosci* 2009; **10**: 861–872.
- 13 Ganguly K, Poo M-M. Activity-dependent neural plasticity from bench to bedside. *Neuron* 2013; **80**: 729–741.
- 14 Hebb DO. *The organization of behaviour*. John Wiley and sons inc.: New York, 1949.
- 15 Sacco RL, Kasner SE, Broderick JP, Caplan LR, Connors JJB, Culebras A et al. An updated definition of stroke for the 21st century: a statement for healthcare professionals from the American Heart Association/American Stroke Association. *Stroke* 2013; **44**: 2064–2089.
- 16 Cooke SF, Bliss TVP. Plasticity in the human central nervous system. *Brain* 2006; **129**: 1659–1673.
- 17 Ogawa S, Lee TM, Nayak AS, Glynn P. Oxygenation-sensitive contrast in magnetic resonance image of rodent brain at high magnetic fields. *Magn Reson Med* 1990; **14**: 68–78.
- 18 Hämäläinen M, Hari R, Ilmoniemi RJ, Knuutila J, Lounasmaa O V. Magnetoencephalography--theory, instrumentation, and applications to noninvasive studies of the working human brain. *Rev Mod Phys* 1993; **65**: 413–497.
- 19 Haith AM, Krakauer JW. *Theoretical models of motor control and motor learning*. 1st editio. Tylor and Francis group, 2013.
- 20 Krakauer JW, Carmichael ST, Corbett D, Wittenberg GF. Getting neurorehabilitation right: what can be learned from animal models? *Neurorehabil Neural Repair* 2012; **26**: 923–931.
- 21 Grefkes C, Fink GR. Reorganization of cerebral networks after stroke: New insights from neuroimaging with connectivity approaches. *Brain* 2011; **134**: 1264–1276.
- 22 Feldman DE, Brecht M. Map plasticity in somatosensory cortex. *Science* 2005; **310**: 810–815.
- 23 Rabiller G, He JW, Nishijima Y, Wong A, Liu J. Perturbation of brain oscillations after ischemic stroke: A potential biomarker for post-stroke function and therapy. *Int J Mol Sci* 2015; **16**: 25605–25640.
- 24 Heiss W-D. The ischemic penumbra: correlates in imaging and implications for treatment of ischemic stroke. The Johann Jacob Wepfer award 2011. *Cerebrovasc Dis* 2011; **32**: 307–320.
- 25 Kwakkel G, Lannin NA, Borschmann K, English C, Ali M, Churilov L et al. Standardized measurement of sensorimotor recovery in stroke trials: Consensus-based core recommendations from the Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2017; **12**: 451–461.

- 26 Buma F, Kwakkel G, Ramsey N. Understanding upper limb recovery after stroke. *Restor Neurol Neurosci* 2013; **31**: 707–722.
- 27 Ward NS. Restoring brain function after stroke - bridging the gap between animals and humans. *Nat Rev Neurol* 2017; **13**: 244–255.
- 28 Carey LM, Lamp G, Turville M. The state-of-the-science on somatosensory function and its impact on daily life in adults and older adults, and following stroke: a scoping review. *OTJR Occup Particip Heal* 2016; **36**: 27S-41S.
- 29 Nitsche MA, Paulus W. Excitability changes induced in the human motor cortex by weak transcranial direct current stimulation. *J Physiol* 2000; **527**: 633–639.
- 30 Rothwell JC, Hallett M, Berardelli A, Eisen A, Rossini P, Paulus W. Magnetic stimulation: motor evoked potentials. The International Federation of Clinical Neurophysiology. *Electroencephalogr Clin Neurophysiol Suppl* 1999; **52**: 97–103.
- 31 Mozaffarian D, Benjamin EJ, Go AS, Arnett DK, Blaha MJ, Cushman M et al. Executive summary: heart disease and stroke statistics--2016 update: a report from the american heart association. *Circulation* 2016; **133**: 447–454.
- 32 Lin Y, Schulze V, Brockmeyer M, Parco C, Karathanos A, Heinen Y et al. Endovascular Thrombectomy as a Means to Improve Survival in Acute Ischemic Stroke: A Meta-analysis. *JAMA Neurol* 2019; **76**: 850–854.
- 33 Saver JL, Goyal M, van der Lugt A, Menon BK, Majoie CBLM, Dippel DW et al. Time to treatment with endovascular thrombectomy and outcomes from ischemic stroke: a meta-analysis. *JAMA* 2016; **316**: 1279–1289.
- 34 Saver JL. Time is brain--quantified. *Stroke* 2006; **37**: 263–266.
- 35 Wardlaw JM, Murray V, Berge E, del Zoppo G, Sandercock P, Lindley RL et al. Recombinant tissue plasminogen activator for acute ischaemic stroke: an updated systematic review and meta-analysis. *Lancet* 2012; **379**: 2364–2372.
- 36 Hussain M, Moussavi M, Korya D, Mehta S, Brar J, Chahal H et al. Systematic Review and Pooled Analyses of Recent Neurointerventional Randomized Controlled Trials: Setting a New Standard of Care for Acute Ischemic Stroke Treatment after 20 Years. *Interv Neurol* 2016; **5**: 39–50.
- 37 Church EW, Gundersen A, Glantz MJ, Simon SD. Number needed to treat for stroke thrombectomy based on a systematic review and meta-analysis. *Clin Neurol Neurosurg* 2017; **156**: 83–88.
- 38 Langhorne P, Bernhardt J, Kwakkel G. Stroke rehabilitation. *Lancet* 2011; **377**: 1693–1702.
- 39 Winters C. *Reactive neurobiological recovery after ischemic stroke, prognosis and intervention*. 2018.
- 40 Kwakkel G, Kollen B, Lindeman E. Understanding the pattern of functional recovery after stroke: facts and theories. *Restor Neurol Neurosci* 2004; **22**: 281–299.
- 41 Newman M. The process of recovery after hemiplegia. *Stroke* 1972; **3**: 702–710.
- 42 Lawrence ES, Coshall C, Dundas R, Stewart J, Rudd AG, Howard R et al. Estimates of the prevalence of acute stroke impairments and disability in a multiethnic population. *Stroke* 2001; **32**: 1279–1284.
- 43 Bernhardt J, Hayward KS, Kwakkel G, Ward NS, Wolf SL, Borschmann K et al. Agreed definitions and a shared vision for new standards in stroke recovery research: The Stroke Recovery and Rehabilitation Roundtable taskforce. *Int J Stroke* 2017; **12**: 444–450.
- 44 van Kordelaar J, van Wegen EEH, Nijland RHM, Daffertshofer A, Kwakkel G. Understanding adaptive motor control of the paretic upper limb early poststroke: the EXPLICIT-stroke program. *Neurorehabil Neural Repair* 2013; **27**: 854–863.
- 45 van Kordelaar J, van Wegen E, Kwakkel G. Impact of time on quality of motor control of the paretic upper limb after stroke. *Arch Phys Med Rehabil* 2014; **95**: 338–344.
- 46 Kwakkel G, Van Wegen E, Burridge JH, Winstein CJ, Van Dokkum L, Alt Murphy M et al. Standardized measurement of quality of upper limb movement after stroke: Consensus-based core recommendations from the Second Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2019; **14**: 783–791.

- 47 Cortes JC, Goldsmith J, Harran MD, Xu J, Kim N, Schambra HM *et al.* A Short and Distinct Time Window for Recovery of Arm Motor Control Early After Stroke Revealed With a Global Measure of Trajectory Kinematics. *Neurorehabil Neural Repair* 2017; **31**: 552–560.
- 48 Veerbeek J, Van Wegen E, Van Peppen R, Van Der Wees PJ, Hendriks E, Rietberg M *et al.* What is the evidence for physical therapy poststroke? A systematic review and meta-analysis. *PLoS One* 2014; **9**: e87987.
- 49 Dietz V, Sinkjaer T. Spastic movement disorder: impaired reflex function and altered muscle mechanics. *Lancet Neurol* 2007; **6**: 725–733.
- 50 van der Krogt HJM, Meskers CGM, de Groot JH, Klomp A, Arendzen JH. The gap between clinical gaze and systematic assessment of movement disorders after stroke. *J Neuroeng Rehabil* 2012; **9**: 61.
- 51 Meskers CGM, Schouten AC, de Groot JH, de Vlucht E, van Hilten BJJ, van der Helm FCT *et al.* Muscle weakness and lack of reflex gain adaptation predominate during post-stroke posture control of the wrist. *J Neuroeng Rehabil* 2009; **6**: 29.
- 52 Heiss W-D, Zaro Weber O. Validation of MRI determination of the Penumbra by PET measurements in ischemic stroke. *J Nucl Med* 2017; **58**: 187–193.
- 53 Schabitz W-R, Steigleder T, Cooper-Kuhn CM, Schwab S, Sommer C, Schneider A *et al.* Intravenous brain-derived neurotrophic factor enhances poststroke sensorimotor recovery and stimulates neurogenesis. *Stroke* 2007; **38**: 2165–2172.
- 54 Ploughman M, Windle V, MacLellan CL, White N, Dore JJ, Corbett D. Brain-derived neurotrophic factor contributes to recovery of skilled reaching after focal ischemia in rats. *Stroke* 2009; **40**: 1490–1495.
- 55 Wieloch T, Nikolich K. Mechanisms of neural plasticity following brain injury. *Curr Opin Neurobiol* 2006; **16**: 258–264.
- 56 Chopp M, Zhang ZG, Jiang Q. Neurogenesis, angiogenesis, and MRI indices of functional recovery from stroke. *Stroke* 2007; **38**: 827–831.
- 57 Heiss W-D. Contribution of Neuro-Imaging for Prediction of Functional Recovery after Ischemic Stroke. *Cerebrovasc Dis* 2017; **44**: 266–276.
- 58 Zhao S, Zhao M, Xiao T, Jolkkonen J, Zhao C. Constraint-induced movement therapy overcomes the intrinsic axonal growth-inhibitory signals in stroke rats. *Stroke* 2013; **44**: 1698–1705.
- 59 Zeiler SR, Krakauer JW. The interaction between training and plasticity in the poststroke brain. *Curr Opin Neurol* 2013; **26**: 609–616.
- 60 Shatz CJ. The developing brain. *Sci Am* 1992; **267**: 60–67.
- 61 Welker E, Rao SB, Dorfl J, Melzer P, van der Loos H. Plasticity in the barrel cortex of the adult mouse: effects of chronic stimulation upon deoxyglucose uptake in the behaving animal. *J Neurosci* 1992; **12**: 153–170.
- 62 Castro-Alamancos MA. Dynamics of sensory thalamocortical synaptic networks during information processing states. *Prog Neurobiol* 2004; **74**: 213–247.
- 63 Winters C, Van Wegen EEH, Daffertshofer A, Kwakkel G. Generalizability of the proportional recovery model for the upper extremity after an ischemic stroke. *Neurorehabil Neural Repair* 2015; **29**: 614–622.
- 64 van der Vliet R, Selles RW, Andrinopoulou E-R, Nijland R, Ribbers GM, Frens MA *et al.* Predicting upper limb motor impairment recovery after stroke: a mixture model. *Ann Neurol* 2020; **87**: 383–393.
- 65 Winters C, Kwakkel G, Van Wegen EEH, Nijland RHM, Veerbeek JM, Meskers CGM. Moving stroke rehabilitation forward: the need to change research. *NeuroRehabilitation* 2018; **43**: 19–30.
- 66 Nijland RHM, van Wegen EEH, Harmeling-van der Wel BC, Kwakkel G. Presence of finger extension and shoulder abduction within 72 hours after stroke predicts functional recovery: early prediction of functional outcome after stroke: the EPOS cohort study. *Stroke* 2010; **41**: 745–750.
- 67 Stinear C. Prediction of recovery of motor function after stroke. *Lancet Neurol* 2010; **9**: 1228–1232.
- 68 Winters C, Kwakkel G, Nijland R, Van Wegen E. When does return of voluntary finger extension occur post-stroke? A prospective cohort study. *PLoS One* 2016; **11**: e0160528.

- 69 Prabhakaran S, Zarahn E, Riley C, Speizer A, Chong JY, Lazar RM et al. Inter-individual variability in the capacity for motor recovery after ischemic stroke. *Neurorehabil Neural Repair* 2008; **22**: 64–71.
- 70 Smith MC, Byblow WD, Barber PA, Stinear CM. Proportional recovery from lower limb motor impairment after stroke. *Stroke* 2017; **48**: 1400–1403.
- 71 Veerbeek, Winters C, Van Wegen EEH, Kwakkel G. Is the proportional recovery rule applicable to the lower limb after a first-ever ischemic stroke? *PLoS One* 2018; **13**: e0189279.
- 72 Lazar RM, Minzer B, Antonello D, Festa JR, Krakauer JW, Marshall RS. Improvement in aphasia scores after stroke is well predicted by initial severity. *Stroke* 2010; **41**: 1485–1488.
- 73 Marchi NA, Ptak R, Di Pietro M, Schnider A, Guggisberg AG. Principles of proportional recovery after stroke generalize to neglect and aphasia. *Eur J Neurol* 2017; **24**: 1084–1087.
- 74 Winters C, Van Wegen EEH, Daffertshofer A, Kwakkel G. Generalizability of the maximum proportional recovery rule to visuospatial neglect early poststroke. *Neurorehabil Neural Repair* 2017; **31**: 334–342.
- 75 Tyson SF, Hanley M, Chillala J, Selley AB, Tallis RC. Sensory loss in hospital-admitted people with stroke: characteristics, associated factors, and relationship with function. *Neurorehabil Neural Repair* 2008; **22**: 166–172.
- 76 Connell LA, Tyson SF. Measures of sensation in neurological conditions: a systematic review. *Clin Rehabil* 2012; **26**: 68–80.
- 77 Carey LM, Matyas TA, Baum C. Effects of somatosensory impairment on participation after stroke. *Am J Occup Ther* 2018; **72**: 1–10.
- 78 Meyer S, De Bruyn N, Krumlinde-Sundholm L, Peeters A, Feys H, Thijs V et al. Associations between sensorimotor impairments in the upper limb at 1 week and 6 months after stroke. *J Neurol Phys Ther* 2016; **40**: 186–195.
- 79 Meyer S, De Bruyn N, Lafosse C, Van Dijk M, Michielsen M, Thijs L et al. Somatosensory impairments in the upper limb poststroke: distribution and association with motor function and visuospatial neglect. *Neurorehabil Neural Repair* 2016; **30**: 731–742.
- 80 Boyd LA, Hayward KS, Ward NS, Stinear CM, Rosso C, Fisher RJ et al. Biomarkers of stroke recovery: consensus-based core recommendations from the stroke recovery and rehabilitation roundtable. *Int J Stroke* 2017; **12**: 480–493.
- 81 Bembenek JP, Kurczyk K, Karli Nski M, Czlonkowska A. The prognostic value of motor-evoked potentials in motor recovery and functional outcome after stroke - a systematic review of the literature. *Funct Neurol* 2012; **27**: 79–84.
- 82 Rossini PM, Burke D, Chen R, Cohen LG, Daskalakis Z, Di Iorio R et al. Non-invasive electrical and magnetic stimulation of the brain, spinal cord, roots and peripheral nerves: Basic principles and procedures for routine clinical and research application. An updated report from an I.F.C.N. Committee. *Clin Neurophysiol* 2015; **126**: 1071–1107.
- 83 Van Kuijk AA, Pasman JW, Hendricks HT, Zwarts MJ, Geurts ACH. Predicting hand motor recovery in severe stroke: the role of motor evoked potentials in relation to early clinical assessment. *Neurorehabil Neural Repair* 2009; **23**: 45–51.
- 84 Hoonhorst MHJ, Nijland RHM, Van den Berg PJS, Emmelot CH, Kollen BJ, Kwakkel G. Does transcranial magnetic stimulation have an added value to clinical assessment in predicting upper-limb function very early after severe stroke? *Neurorehabil Neural Repair* 2018; **32**: 682–690.
- 85 Stinear CM, Barber PA, Petoe M, Anwar S, Byblow WD. The PREP algorithm predicts potential for upper limb recovery after stroke. *Brain* 2012; **135**: 2527–2535.
- 86 Byblow WD, Stinear CM, Barber PA, Petoe MA, Ackerley SJ. Proportional recovery after stroke depends on corticomotor integrity. *Ann Neurol* 2015; **78**: 848–859.
- 87 Nijboer TCW, Buma FE, Winters C, Vansteensel MJ, Kwakkel G, Ramsey NF et al. No changes in functional connectivity during motor recovery beyond 5 weeks after stroke; A longitudinal resting-state fMRI study. *PLoS One* 2017; **12**: e0178017.
- 88 Buma FE, Lindeman E, Ramsey NF, Kwakkel G. Review: functional neuroimaging Studies of early upper limb recovery after stroke: a systematic review of the literature. *Neurorehabil Neural Repair* 2010; **24**: 589–608.

- 89 Saur D, Lange R, Baumgaertner A, Schraknepper V, Willmes K, Rijntjes M et al. Dynamics of language reorganization after stroke. *Brain* 2006; **129**: 1371–1384.
- 90 Corbetta M, Kincade MJ, Lewis C, Snyder AZ, Sapir A. Neural basis and recovery of spatial attention deficits in spatial neglect. *Nat Neurosci* 2005; **8**: 1603–1610.
- 91 Rossini PM, Altamura C, Ferreri F, Melgari J-M, Tecchio F, Tombini M et al. Neuroimaging experimental studies on brain plasticity in recovery from stroke. *Eura Medicophys* 2007; **43**: 241–254.
- 92 Carey LM, Abbott DF, Harvey MR, Puce A, Seitz RJ, Donnan GA. Relationship between touch impairment and brain activation after lesions of subcortical and cortical somatosensory regions. *Neurorehabil Neural Repair* 2011; **25**: 443–457.
- 93 Nudo RJ. Mechanisms for recovery of motor function following cortical damage. *Curr Opin Neurobiol* 2006; **16**: 638–644.
- 94 Cramer SC. Repairing the human brain after stroke: I. Mechanisms of spontaneous recovery. *Ann. Neurol.* 2008; **63**: 272–287.
- 95 Manganotti P, Hallett M, Leocani L, Toro C, Grafman J, Zhuang P. Event-related desynchronization (ERD) in the alpha frequency during development of implicit and explicit learning. *Electroencephalogr Clin Neurophysiol* 2002; **102**: 374–381.
- 96 Campfens SF, Schouten AC, van Putten MJAM, van der Kooij H. Quantifying connectivity via efferent and afferent pathways in motor control using coherence measures and joint position perturbations. *Exp brain Res* 2013; c: 141–153.
- 97 Burghaus L, Hilker R, Dohmen C, Bosche B, Winhuisen L, Galdiks N et al. Early electroencephalography in acute ischemic stroke: Prediction of a malignant course? *Clin Neurol Neurosurg* 2007; **109**: 45–49.
- 98 Sheorajpanday RVA, Nagels G, Weeren AJTM, van Putten MJAM, De Deyn PP. Quantitative EEG in ischemic stroke: correlation with functional status after 6 months. *Clin Neurophysiol* 2011; **122**: 874–883.
- 99 Van Putten MJAM, Tavy DLJ. Continuous quantitative EEG monitoring in hemispheric stroke patients using the brain symmetry index. *Stroke* 2004; **35**: 2489–2492.
- 100 Stagg CJ, Antal A, Nitsche MA. Physiology of transcranial Direct Current Stimulation. *J ECT* 2018; **34**: 144–152.
- 101 Nitsche MA, Fricke K, Henschke U, Schlitterlau A, Liebetanz D, Lang N et al. Pharmacological modulation of cortical excitability shifts induced by transcranial direct current stimulation in humans. *J Physiol* 2003; **553**: 293–301.
- 102 Clark VP, Coffman BA, Trumbo MC, Gasparovic C. Transcranial direct current stimulation (tDCS) produces localized and specific alterations in neurochemistry: a Magnetic resonance spectroscopy study. *Neurosci Lett* 2011; **500**: 67–71.
- 103 Monte-Silva K, Kuo MF, Hessenthaler S, Fresnoza S, Liebetanz D, Paulus W et al. Induction of late LTP-like plasticity in the human motor cortex by repeated non-invasive brain stimulation. *Brain Stimul* 2013; **6**: 424–432.
- 104 Batsikadze G, Moliadze V, Paulus W, Kuo MF, Nitsche MA. Partially non-linear stimulation intensity-dependent effects of direct current stimulation on motor cortex excitability in humans. *J Physiol* 2013; **591**: 1987–2000.
- 105 Lisman JE. Three Ca<sup>2+</sup> levels affect plasticity differently: the LTP zone, the LTD zone and no man's land. *J Physiol* 2001; **532**: 285.
- 106 Malenka RC, Bear MF. LTP and LTD: an embarrassment of riches. *Neuron* 2004; **44**: 5–21.
- 107 Stagg CJ, Best JG, Stephenson MC, O'Shea J, Wylezinska M, Kincses ZT et al. Polarity-sensitive modulation of cortical neurotransmitters by transcranial stimulation. *J Neurosci* 2009; **29**: 5202–5206.
- 108 Stagg CJ, Bestmann S, Constantinescu AO, Moreno LM, Allman C, Mekle R et al. Relationship between physiological measures of excitability and levels of glutamate and GABA in the human motor cortex. *J Physiol* 2011; **589**: 5845–5855.
- 109 Bachtiar V, Near J, Johansen-Berg H, Stagg CJ. Modulation of GABA and resting state functional connectivity by transcranial direct current stimulation. *Elife* 2015; **4**: e08789.

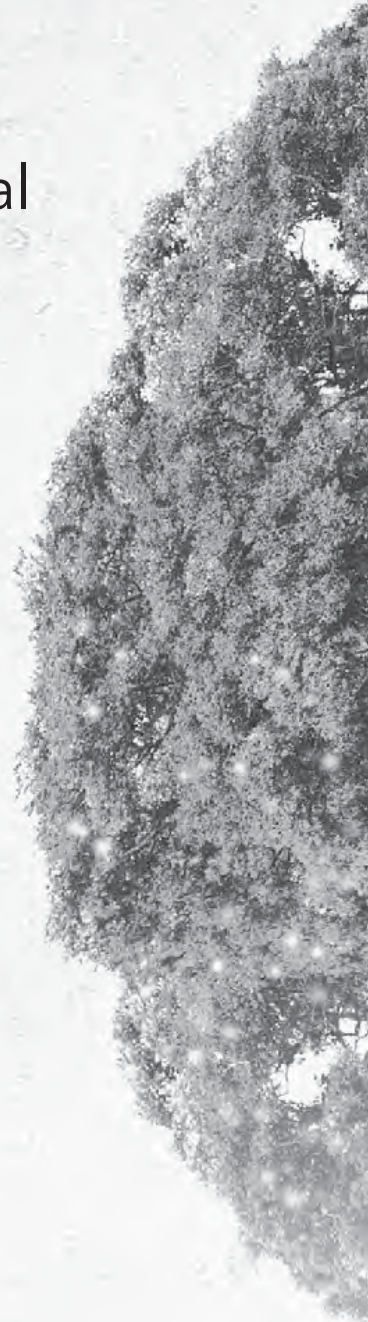
- 110 Nitsche MA, Muller-Dahlhaus F, Paulus W, Ziemann U. The pharmacology of neuroplasticity induced by non-invasive brain stimulation: building models for the clinical use of CNS active drugs. *J Physiol* 2012; **590**: 4641–4662.
- 111 Fritsch B, Reis J, Martinowich K, Schambra HM, Ji Y, Cohen LG et al. Direct current stimulation promotes BDNF-dependent synaptic plasticity: Potential implications for motor learning. *Neuron* 2010; **66**: 198–204.
- 112 Podda MV, Cocco S, Mastrodonato A, Fusco S, Leone L, Barbati SA et al. Anodal transcranial direct current stimulation boosts synaptic plasticity and memory in mice via epigenetic regulation of Bdnf expression. *Sci Rep* 2016; **6**: 22180.
- 113 Kowiański P, Lietzau G, Czuba E, Waśkow M, Steliga A, Moryś J. BDNF: a key factor with multipotent impact on brain signaling and synaptic plasticity. *Cell Mol Neurobiol* 2018; **38**: 579–593.
- 114 Elsner B, Kugler J, Pohl M, Mehrholz J. Transcranial direct current stimulation (tDCS) for improving function and activities of daily living in patients after stroke. *Cochrane database Syst Rev* 2013; **11**: CD009645.
- 115 Elsner B, Kugler J, Pohl M, Mehrholz J. Transcranial direct current stimulation (tDCS) for improving activities of daily living, and physical and cognitive functioning, in people after stroke. *Cochrane database Syst Rev* 2016; **3**: CD009645.
- 116 Elsner B, Kwakkel G, Kugler J, Mehrholz J. Transcranial direct current stimulation (tDCS) for improving capacity in activities and arm function after stroke: a network meta-analysis of randomised controlled trials. *J Neuroeng Rehabil* 2017; **14**: 95.
- 117 Marquez J, Van Vliet P, Mcelduff P, Lagopoulos J, Parsons M. Transcranial direct current stimulation (tDCS): Does it have merit in stroke rehabilitation? A systematic review. *Int. J. Stroke*. 2015; **10**: 306–316.
- 118 D'Angelo E. The organization of plasticity in the cerebellar cortex: from synapses to control. *Prog Brain Res* 2014; **210**: 31–58.
- 119 Medina JF, Lisberger SG. Links from complex spikes to local plasticity and motor learning in the cerebellum of awake-behaving monkeys. *Nat Neurosci* 2008; **11**: 1185–1192.
- 120 Schlerf J, Ivry RB, Diedrichsen J. Encoding of sensory prediction errors in the human cerebellum. *J Neurosci* 2012; **32**: 4913–4922.
- 121 Pollok B, Butz M, Gross J, Südmeyer M, Timmermann L, Schnitzler A. Coupling between cerebellar hemispheres: Behavioural, anatomic, and functional data. *Cerebellum* 2006; **5**: 212–219.
- 122 Azevedo FAC, Carvalho LRB, Grinberg LT, Farfel JM, Ferretti REL, Leite REP et al. Equal numbers of neuronal and nonneuronal cells make the human brain an isometrically scaled-up primate brain. *J Comp Neurol* 2009; **513**: 532–541.
- 123 Rampersad SM, Janssen AM, Lucka F, Aydin U, Lanfer B, Lew S et al. Simulating transcranial direct current stimulation with a detailed anisotropic human head model. *IEEE Trans Neural Syst Rehabil Eng* 2014; **22**: 441–452.
- 124 Galea JM, Jayaram G, Ajagbe L, Celnik P. Modulation of cerebellar excitability by polarity-specific noninvasive direct current stimulation. *J Neurosci* 2009; **29**: 9115–9122.
- 125 Schonewille M, Belmeguenai A, Koekoek SK, Houtman SH, Boele HJ, van Beugen BJ et al. Purkinje cell-specific knockout of the protein phosphatase PP2B impairs potentiation and cerebellar motor learning. *Neuron* 2010; **67**: 618–628.
- 126 Schonewille M, Gao Z, Boele HJ, Vinuesa Veloz MF, Amerika WE, Simek AAM et al. Reevaluating the role of LTD in cerebellar motor learning. *Neuron* 2011; **70**: 43–50.
- 127 Caligiore D, Pezzulo G, Baldassarre G, Bostan AC, Strick PL, Doya K et al. Consensus Paper: Towards a Systems-Level View of Cerebellar Function: the Interplay Between Cerebellum, Basal Ganglia, and Cortex. *Cerebellum* 2017; **16**: 203–229.
- 128 Schlerf JE, Galea JM, Spampinato D, Celnik PA. Laterality Differences in Cerebellar-Motor Cortex Connectivity. *Cereb Cortex* 2015; **25**: 1827–1834.
- 129 Medina JF, Lisberger SG. Links from complex spikes to local plasticity and motor learning in the cerebellum of awake-behaving monkeys. *Nat Neurosci* 2008; **11**: 1185–1192.
- 130 Ito M, Yamaguchi K, Nagao S, Yamazaki T. Long-Term Depression as a model of cerebellar plasticity. *Prog Brain Res* 2014; **210**: 1–30.



- 131 Palesi F, De Rinaldis A, Castellazzi G, Calamante F, Muhlert N, Chard D et al. Contralateral cortico-ponto-cerebellar pathways reconstruction in humans in vivo: implications for reciprocal cerebro-cerebellar structural connectivity in motor and non-motor areas. *Sci Rep* 2017; 7: 12841.
- 132 Jayaram G, Tang B, Pallegadda R, Vasudevan EVL, Celnik P, Bastian A. Modulating locomotor adaptation with cerebellar stimulation. *J Neurophysiol* 2012; 107: 2950–2957.
- 133 Wessel MJ, Hummel FC. Non-invasive cerebellar stimulation: a promising approach for stroke recovery? *Cerebellum* 2018; 17: 359–371.
- 134 Carter AR, Chen C, Schwartz PM, Segal R a. Brain-derived neurotrophic factor modulates cerebellar plasticity and synaptic ultrastructure. *J Neurosci* 2002; 22: 1316–1327.
- 135 Geurts ACH, De Haart M, Van Nes IJW, Duysens J. A review of standing balance recovery from stroke. *Gait Posture*. 2005; 22: 267–281.
- 136 Morton SM, Bastian AJ. Cerebellar control of balance and locomotion. *Neuroscientist* 2004; 10: 247–259.
- 137 Thach WT, Bastian AJ. Role of the cerebellum in the control and adaptation of gait in health and disease. *Prog Brain Res* 2004; 143: 353–366.
- 138 Stagg CJ, Jayaram G, Pastor D, Kincses ZT, Matthews PM, Johansen-Berg H. Polarity and timing-dependent effects of transcranial direct current stimulation in explicit motor learning. *Neuropsychologia* 2011; 49: 800–804.
- 139 Nudo RJ, Plautz EJ, Frost SB. Role of adaptive plasticity in recovery of function after damage to motor cortex. *Muscle Nerve* 2001; 24: 1000–1019.
- 140 Walker MF, Hoffmann TC, Brady MC, Dean CM, Eng JJ, Farrin AJ et al. Improving the development, monitoring and reporting of stroke rehabilitation research: Consensus-based core recommendations from the Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2017; 12: 472–479.
- 141 Corbett D, Carmichael ST, Murphy TH, Jones TA, Schwab ME, Jolkkonen J et al. Enhancing the alignment of the preclinical and clinical stroke recovery research pipeline: Consensus-based core recommendations from the Stroke Recovery and Rehabilitation Roundtable translational working group. *Int J Stroke* 2017; 12: 462–471.

# PART 1

Spontaneous neurobiological  
recovery after stroke









# Chapter 2

Is recovery of  
somatosensory  
impairment conditional  
for upper limb motor  
recovery early after  
stroke?

Sarah B. Zandvliet  
Gert Kwakkel  
Rinske H.M. Nijland  
Erwin E.H. van Wegen  
Carel G.M. Meskers

*Neurorehabilitation and Neural Repair* 2020;  
34(5): 403–416

## ABSTRACT

**Background:** Spontaneous recovery early after stroke is most evident during a time-sensitive window of heightened neuroplasticity, known as spontaneous neurobiological recovery. It is unknown whether post-stroke upper limb motor and somatosensory impairment both reflect spontaneous neurobiological recovery or if somatosensory impairment and/or recovery influences motor recovery.

**Methods:** Motor (Fugl-Meyer motor assessment of the upper extremity [FM-UE]) and somatosensory impairments (Erasmus modification of the Nottingham sensory assessment [EmNSA-UE]) were measured in 215 patients within 3, at 5, 12 and 26 weeks after a first-ever ischemic stroke. The longitudinal association between FM-UE and EmNSA-UE was examined in patients with motor and somatosensory impairments (FM-UE $\leq$ 60 and EmNSA-UE $\leq$ 37) at baseline.

**Results:** Ninety-four patients were included in the longitudinal analysis. EmNSA-UE increased significantly up to 12 weeks post-stroke. The longitudinal association between motor and somatosensory impairment disappeared when correcting for progress of time, and was not significantly different for patients with severe baseline somatosensory impairment. Patients with a FM-UE score  $\geq$ 18 at 26 weeks (N=55) showed a significant positive association between motor and somatosensory impairments, irrespective of progress of time.

**Conclusions:** Progress of time, as a reflection of spontaneous neurobiological recovery is an important factor that drives recovery of upper limb motor as well as somatosensory impairments in the first 12 weeks post-stroke. Severe somatosensory impairment at baseline does not directly compromise motor recovery. The study rather suggests that spontaneous recovery of somatosensory impairment is a prerequisite for full motor recovery of the upper paretic limb.



## INTRODUCTION

Somatosensory impairment is common in the acute phase after stroke, with prevalence rates between 34% and 84%<sup>1-4</sup>, and is associated with reduced upper limb motor function, activity and participation post-stroke and relate to increased hospital length of stay<sup>3-6</sup>. The relation between motor and somatosensory impairments in the first 3 months post-stroke may reflect parallel recovery in both modalities, driven by a common underlying neurobiological mechanism that occurs in a time-sensitive window of heightened neuroplasticity early after stroke<sup>7-10</sup>, known as spontaneous neurobiological recovery<sup>8,9</sup>. A number of clinical observational studies however suggest that severe somatosensory impairment may hamper motor recovery post-stroke<sup>11,12</sup>. This relationship may specifically be explained by the importance of somatosensory input for fine motor skills of the upper limb<sup>12</sup>.

Previous prospective studies found that spontaneous neurobiological recovery as reflected by progress of time alone, is the most significant covariate for explaining the recovery pattern of neurological impairments in the first 8 to 10 weeks post-stroke<sup>7</sup>. In addition, a number of observational studies indicated that spontaneous neurobiological recovery is proportional to initial upper limb<sup>13-15</sup>, lower limb<sup>16,17</sup>, and somatosensory impairments<sup>18,19</sup>, aphasia<sup>20,21</sup> and visual spatial neglect (VSN)<sup>21,22</sup>, with a recovery range between 64<sup>16,19</sup> up to 97%<sup>22</sup>. Patients who failed to show spontaneous recovery of VSN after a first-ever ischemic right hemispheric stroke, also have a high probability to fail recovery on other affected modalities such as motor impairment of the upper paretic limb (i.e. so-called non-recoverers of spontaneous neurobiological recovery)<sup>22</sup>. Nijboer et al. showed that less improvement on the Fugl-Meyer motor assessment of the upper extremity (FM-UE) was independently associated with more severe VSN in the first 10 weeks post-stroke, suggesting a suppressive effect of neglect on upper limb motor recovery within the time window of spontaneous neurobiological recovery<sup>23</sup>. Finally, evidence was found that patients who did not show a pattern of spontaneous neurobiological recovery in their lower limb, are also not likely to show upper limb recovery within the first 6 months post-stroke<sup>16</sup>. These results suggest that post-stroke recovery is driven by common underlying processes reflected by spontaneous neurobiological recovery spanning multiple modalities<sup>9,13-16,18-22</sup>. Multiple processes such as salvation of penumbral tissue<sup>24</sup>, upregulation of growth promoting factors, gene-dependent enhancement of angiogenesis<sup>25</sup> and alleviation of diaschisis<sup>24</sup>, are mentioned as factors that may drive spontaneous neurobiological recovery. Unfortunately, above mentioned mechanisms are still poorly understood and no causal marker has yet been identified that can accurately predict who will or will not show spontaneous neurobiological recovery early after stroke<sup>8,9,26,27</sup>.

Meyer et al. previously showed, in a cross-sectional study in 122 patients within the first 6 months post-stroke, that motor and somatosensory impairments are low to moderately correlated ( $r=0.22-0.61$ )<sup>4</sup>. To further disentangle the relationship between motor and somatosensory recovery, i.e. whether both can be explained from general mechanisms of spontaneous neurobiological recovery or if somatosensory impairment and/or recovery influences motor recovery, a longitudinal study is required. In addition, the absence of somatosensory input could compromise experience-dependent plasticity, which underlies the remodeling of neural circuits and could therefore impair the development of new motor programs after stroke<sup>28-31</sup>. In this latter situation, one expects a failure in recovery of somatosensory impairment to be significantly associated with less motor recovery of the upper paretic limb.

In the present study, we aimed to describe the time course of somatosensory recovery and to analyze the longitudinal association between motor and somatosensory impairments in the first 6 months post-stroke. We examined if the association between motor and somatosensory impairments remained after adjusting for progress of time, as a reflection of spontaneous neurobiological recovery<sup>7</sup>, and whether this longitudinal association was different in patients with an initially severe baseline level of somatosensory impairment when compared to those with a mild to moderate sensory impairment in the first week post-stroke. Finally, we aimed to investigate whether the association between motor and somatosensory impairments depend on the presence of motor recovery of the upper paretic limb. For this latter aim we investigated the difference, between patients who showed motor recovery (i.e., recoverers) compared with those who did not show spontaneous motor recovery of the upper limb (i.e., non-recoverers) in the first 6 months post-stroke.

## MATERIALS AND METHODS

Data were derived from three longitudinal studies, i.e. the EXPLICIT<sup>32</sup>, EXPLORE-stroke and 4D-EEG cohorts, with a total of 215 patients. The EXPLICIT randomized controlled trial (RCT) investigated the effects of a modified constraint-induced movement therapy (mCIMT) and EMG-triggered neuro-muscular stimulation (EMG-NMS) on stroke recovery mechanisms compared to usual care (Trial NL1366, NTR1424). Patients were included within 3 weeks post-stroke and assessed weekly during the first 5 weeks and then at 8, 12 and 26 weeks post-stroke<sup>33</sup>. Voluntary finger extension was used to stratify patients into a group with a favorable prognosis for upper limb motor recovery, who received mCIMT or usual care, and a group with an unfavorable prognosis, who received EMG-NMS or usual care. Neither mCIMT nor EMG-NMS significantly influenced upper limb motor

recovery in terms of FM-UE at any time point in the first 6 months post-stroke<sup>32</sup>. Hence, the present study used data of the total sample. Patients enrolled the EXPLORE-stroke or 4D-EEG cohort studies all received usual care following the current Dutch guidelines of physiotherapy<sup>34</sup>.

EXPLORE-stroke and 4D-EEG (Trial NL4084, NTR4221) were longitudinal observational cohort studies that both assessed clinical scales as well as neurophysiological parameters in a repetitive manner to improve prediction models and to enhance understanding of functional recovery after stroke. In line with recent recommendations<sup>8</sup>, clinical assessments in the EXPLORE-stroke and 4D-EEG studies were made at fixed times post-stroke, i.e., within 3 weeks and at 5, 12 and 26 weeks post-stroke. Patients in the 4D-EEG study were additionally assessed at 8 weeks post-stroke.

Within the aforementioned cohorts, the following inclusion criteria were used: (1) having experienced a first-ever, ischemic hemispheric stroke, verified by CT and/or MRI scan less than 3 weeks before inclusion; (2) having an upper limb paresis as defined by a national institutes of health stroke scale (NIHSS) score of 1 or more; (3) being aged between 18 and 80 years; (4) having no severe cognitive deficits (mini mental state examination of at least 19 points)<sup>35,36</sup>; (5) being able to sit for 30 seconds without support; (6) having no orthopedic limitations of the upper limb; (7) having no pre-existing neurological condition. All procedures were in accordance with the declaration of Helsinki and were approved by the medical ethics committees of Leiden university medical center (EXPLICIT: NL21396.058.08, EXPLORE-stroke: NL39323.058.12) or VU university medical center (4D-EEG: NL47079.029.14). All participants gave their written informed consent.

### **Measuring somatosensory impairment and determining baseline level of impairment**

Somatosensory impairment of the upper extremity was assessed using the Erasmus modification of the Nottingham sensory assessment (EmNSA)<sup>37</sup>. The intra- and interrater reliability of the EmNSA for the upper limb are predominantly good to excellent ( $\kappa=0.62-1.00$  intra- and  $\kappa=0.48-1.00$  inter-rater reliability) for patients with intracranial disorders<sup>37</sup>. The EmNSA uses a 3-point ordinal scale and offers a reliable somatosensory assessment of the upper and lower limb for patients with intracranial disorders. The testing procedure includes a pinprick test to assess tactile sensation, sharp-blunt discrimination to assess pain sensation and measuring proprioception to assess gnostic sensibility. The maximum score of the EmNSA for the upper extremity (EmNSA-UE) is 40 points. A score of 39 points

or lower has been described as a somatosensory impairment<sup>37</sup>, since the measurement error of the EmNSA-UE has not been established, we considered a baseline score below 38 points as somatosensory impairment, accounting for a measurement error of 5%.

#### *Defining baseline level of impairment following EmNSA-UE*

Patients were categorized into high and a low baseline scores on the EmNSA-UE to differentiate between severe and moderate somatosensory impairments (baseline EmNSA-UE level). To distinguish between these groups, a dichotomous variable was constructed based on the NIHSS item score of somatosensory impairment at 26 weeks post-stroke, distinguishing between having no somatosensory impairment (0 points) and having a somatosensory impairment (1 or 2 points) as the state variable in the receiver operating characteristic (ROC) curve<sup>38</sup>. The cut-off for low and high baseline scores on the EmNSA-UE within 3 weeks post-stroke was determined by inspecting the ROC curve, in which an optimum between sensitivity and specificity was sought, prioritizing sensitivity.

#### **Measuring motor impairment, determining grouping variables: baseline level of impairment and recovery patterns**

Motor impairment of the upper limb was measured with the Fugl-Meyer motor assessment of the upper extremity (FM-UE)<sup>39,40</sup>. To account for a 6 point measurement error, patients were considered to have a motor impairment when the baseline score was 60 (out of the 66) points or less<sup>41</sup>.

#### *Defining baseline level of impairment following FM-UE*

The baseline level for severe motor impairment was set at a cut-off of 18 points on the FM-UE<sup>9,13,14</sup>. The 18-point FM-UE cut-off derived from Winters et al. was checked for suitability for the current study by constructing a ROC-curve. The NIHSS item on motor impairment of the affected upper limb was used to construct the state variable, after which the same steps were applied as those described for somatosensory impairment level.

#### *Defining the recovery pattern subgroups, i.e. recoverers and non-recoverers following FM-UE*

In case of a severe baseline level of motor impairment, a further distinction in motor recovery pattern subgroups was made. This distinction between 'recoverers' and 'non-recoverers' was made based on whether or not a patient showed clinically relevant improvement<sup>41</sup> on the FM-UE over time as an indication of spontaneous neurobiological recovery, and was defined as:

- Non-recoverers: FM-UE score <18 at baseline, <6 points improvement or FM-UE score <18 points at 26 weeks post-stroke
- Recoverers: FM-UE score <18 points at baseline with  $\geq 6$  points improvement resulting in  $\geq 18$  points at 26 weeks post-stroke

### Measuring covariates

Covariates which are assumed to affect or are associated with sensorimotor recovery of the upper limb<sup>14,23,42,43</sup>, were considered as possible confounders in the longitudinal association between motor and somatosensory recovery. These were: 1) age; 2) affected hemisphere; 3) comorbidities, measured with the cumulative illness rating score (CIRS)<sup>44</sup>; 4) visuospatial neglect, assessed with a single-target letter cancellation test (LCT)<sup>45</sup>. Patients were instructed to mark all O's on an A4 sheet, which was aligned to the patient's sagittal midline. The sheet showed 20 O's on both sides of the midline, mixed with random letters. The marked O's in the contralesional visual field were counted; 5) Motor impairment of the lower limb, measured with the motricity index of the lower extremity (MI-LE)<sup>46</sup>; 6) stroke severity, longitudinally measured with the national institutes of health stroke scale (NIHSS)<sup>47</sup>. This scale evaluates the severity of possibly affected modalities after stroke. To account for overlaps with FM-UE, EmNSA-UE, LCT and MI-LE, a NIHSS-adapt variable was constructed by leaving out items that measure upper and lower limb motor impairment, limb ataxia, somatosensory impairment, extinction and inattention (items 5a, 5b, 6a, 6b, 7, 8 and 11).

### Statistical analysis

The time course of somatosensory recovery was described using a mixed model analysis, with the EmNSA-UE scores from baseline until 26 weeks post-stroke for patients with both a motor and sensory impairment, based on abovementioned cut off values. The longitudinal association between motor (FM-UE) and somatosensory impairments (EmNSA-UE) was analyzed using a second association model.

To evaluate if this association was robust for confounders, we examined the influence of covariates: age, lesion side, CIRS, NIHSS-adapt, LCT, and MI-LE. An adjustment for time was subsequently made to determine if the association was partly independent of progress of time and therefore of spontaneous neurobiological recovery. We further evaluated the interaction effect of severity of somatosensory impairment at baseline, on the longitudinal association of recovery between motor and somatosensory impairments.

We investigated whether the association between motor and somatosensory recovery differed for patients with a low versus a high baseline score on the FM-UE and for recoverers versus non-recoverers. We investigated this using two different methods. For the first method we constructed dichotomous grouping variables based on the baseline FM-UE level and the FM-UE recovery pattern subgroups, as described above i.e. recoverers and non-recoverers. Interaction terms between the baseline FM-UE level and EmNSA-UE and between FM-UE recovery patterns and EmNSA-UE were added and evaluated for statistical significance. Secondly, as an alternative for defining grouping variables, an alternative model was used in which within- and between-subject effects are separated<sup>48</sup>. This, so called, hybrid model enables a direct distinction between factors relating to differences in recovery within a patient over time and factors relating to differences in recovery between patients<sup>48</sup>. For this purpose, we calculated the mean EmNSA-UE score for each individual patient, representing the between-subject part of the association, and the EmNSA-UE scores per measurement moment minus the patient's mean score, representing the within-subject part. The association between FM-UE and EmNSA-UE was reanalyzed, estimating two separate beta-coefficients to represent the within- and between-subject parts of the association.

To correct for dependency between measurements, a random intercept per patients was used in the models. Residuals were checked for normality by inspection of the probability distributions (q-q plots) and histograms. Significance level was set at a two-tailed Alpha of 0.05 for all analyses. All analyses were performed using IBM SPSS statistics version 22 (IBM corporation, Armonk, NY, USA).

## RESULTS

The flowchart of patient inclusion is shown in Figure 2.1. Subject's characteristics are displayed in Table 2.1. Data of 215 patients was collected within the three abovementioned studies between October 2008 and May 2017. Of the 197 patients from which sufficient data was collected, 195 patients had a motor impairment, i.e. an FM-UE score of 60 point or less at baseline. Ninety-five patients (49.0% of the patients with a complete dataset) had an initial somatosensory impairment following the EmNSA-UE score of 37 point or less at baseline. Combining both modalities, data of 94 patients was available to determine the longitudinal association. All residuals of the mixed model analysis were normally distributed.

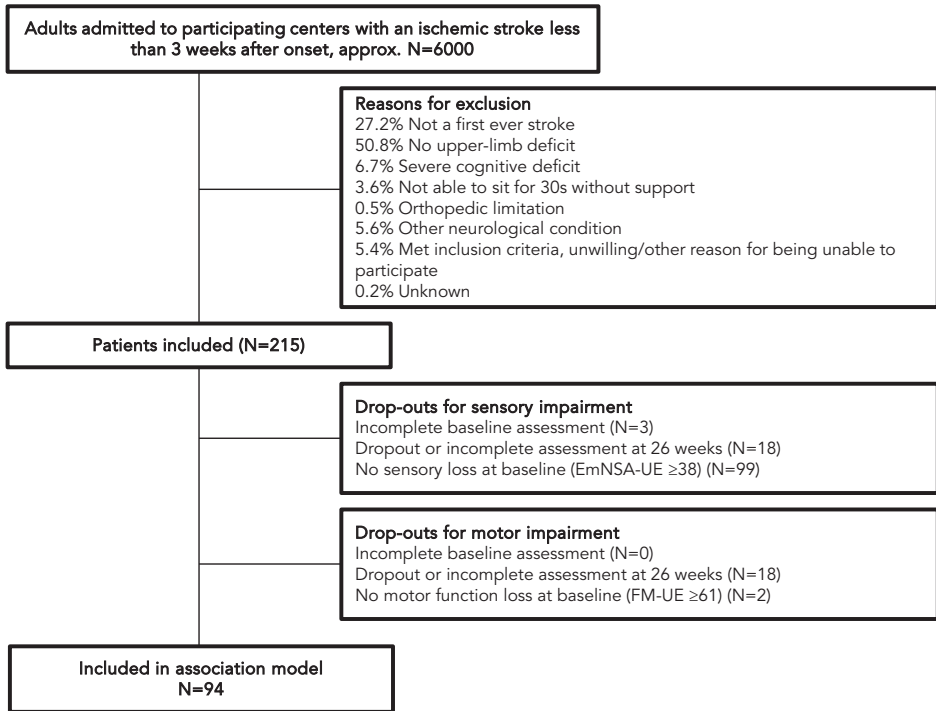


Figure 2.1 | Flowchart of the included patients with a stroke.

Abbreviations: Erasmus modification of the Nottingham sensory assessment of the upper extremity (EmNSA-UE), Fugl-Meyer motor assessment of the upper extremity (FM-UE), number of patients (N).

### Time course of somatosensory recovery

Figure 2.2 shows the individual time courses of motor and somatosensory recovery for the 94 patients included in the association model up to 26 weeks post-stroke. Mean EmNSA-UE increased significantly up to week 12 post-stroke. While no further significant increase was found between 12 and 26 weeks at a group level, there is evidence of change at an individual level, as can be seen in Figure 2.2. Table 2.2 displays the corresponding descriptives and the beta estimates with 95% confidence intervals (CI) and probability values of the association model describing the time course of somatosensory recovery.

### *Longitudinal association of motor and somatosensory impairments and impact of progress of time*

Figure 2.3 shows the recovery of motor and somatosensory impairments expressed as a percentage of the maximum possible recovery on the FM-UE and EmNSA-UE, as



a visual illustration of their relationship. Most patients (N=78) showed relatively more somatosensory than motor recovery, see Figure 2.3. Thirteen patients (14%) showed more motor than somatosensory recovery. Table 2.3A displays the outcome of the association

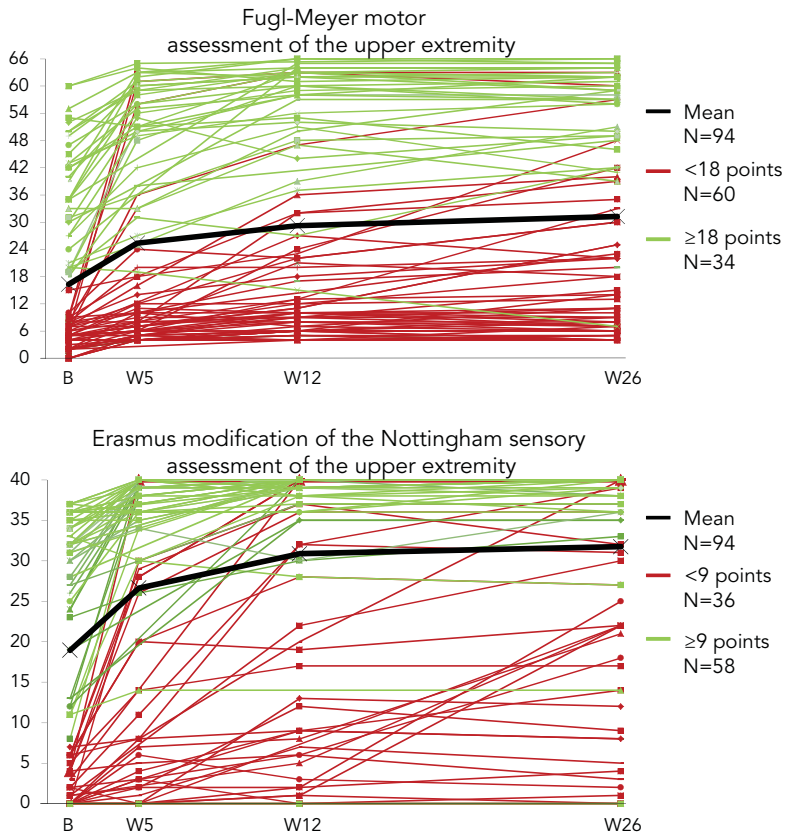
Table 2.1 | Subject's characteristics

Characteristic	All subjects	High baseline motor score	Recoverers	Non-recoverers
	N=94	N=34	N=21	N=39
Time between stroke and baseline measurements (days) <sup>a</sup>	9.6 (4.7)	10.7 (5.0)	10.0 (4.8)	8.3 (4.1)
Age (y) <sup>a</sup>	60.3 (12.5)	60.6 (14.3)	62.1 (10.9)	59.1 (11.8)
Gender, male/female (N) <sup>b</sup>	58/34	21/13	12/9	25/14
Affected hemisphere, left/right/(N) <sup>b</sup>	27/67	10/24	9/12	8/31
Bamford classification, LACI, PACI or TACI (N) <sup>b</sup>	32/53/9	14/18/2	8/11/2	10/24/5
CIRS	2 (2–4)	3.5 (2–5.25)	2 (2–4)	2 (1–4)
NIHSS	9 (5–12)	5 (3.75–7)	8.5 (8–10)	12 (10–13)
LCT at baseline	14 (3–19)	19 (14–20)	17.5 (13.25–20)	4 (0–14)
LCT at 6 months ps	19.5 (17–20)	20 (19–20)	20 (18.25–20)	19 (15–20)
FM-UE at baseline	7 (4–30)	35 (26.25–47.5)	7 (5.5–8.5)	4 (2–5)
FM-UE at 6 months ps	24 (7.75–57)	58.5 (49–62.25)	33 (22–52.5)	7 (5–9)
ARAT at baseline	0 (0–6.5)	16 (6–27.5)	0 (0–0)	0 (0–0)
ARAT at 6 months ps	10.5 (0–43.25)	49.5 (37.75–55)	22 (7–39.5)	0 (0–0)
EmNSA-UE at baseline	24 (2–34)	32 (10.75–36)	32 (4.5–35.5)	6 (0–25)
EmNSA-UE at 6 months ps	39.5 (24.25–40)	40 (36.75–40)	40 (36.5–40)	35 (14–40)
MI-UE at baseline	11 (0–49.5)	58 (47–65)	12.5 (0–28.75)	0 (0–0)
MI-UE at 6 months ps	47 (18–76)	84 (76–92)	65 (47–76)	14 (0–28)
MI-LE at baseline	42 (9–64)	75 (53–100)	42 (28–56.75)	9 (0–23)
MI-LE at 6 months ps	69 (47–89)	100 (77.5–100)	72 (64–75)	43 (37–64)

FM-UE score of 18 points or higher is considered a high baseline score for motor impairment. Patients with an FM-UE score below 18 points were divided into recoverers (FM-UE  $\geq$ 18 points at 26 weeks and at least a 6-point improvement between baseline and 26 weeks post-stroke) and non-recoverers (FM-UE <18 points at 26 weeks post-stroke or failing to show a 6-point improvement between baseline and 26 weeks post-stroke). Unless indicated otherwise the provided scale is ordinal and median and interquartile ranges are displayed. Continuous variable (a); mean and standard deviation are displayed. categorical/nominal variable (b); number of patients is displayed. Years (y), kilogram (kg), meter (m), number of subjects (N), lacunar anterior circulation infarct (LACI), partial anterior circulation infarct (PACI), total anterior circulation infarct (TACI), cumulative illness rating scale (CIRS), national institute of health stroke scale (NIHSS), letter cancellation test (LCT), Fugl-Meyer motor assessment of the upper extremity (FM-UE), action research arm test (ARAT), Erasmus modification of the Nottingham sensory assessment of the upper extremity (EmNSA-UE), motricity index (MI), lower extremity (LE), upper extremity (UE), post-stroke (ps). Baseline value is the first measurement of each subject within 3 weeks post-stroke.

model between motor and somatosensory impairments. FM-UE and EmNSA-UE showed a significant longitudinal association ( $\beta$  intercept=10.91,  $\beta$  EmNSA-UE=0.55,  $P<0.01$ ). The longitudinal association between FM-UE and EmNSA-UE changed, yet remained significant, when adjusting for age, lesion side, CIRS, NIHSS-adapt, LCT and MI-LE, with  $\beta$  intercept=3.42 and  $\beta$  EmNSA-UE=0.21,  $P<0.01$ . The corresponding confidence intervals are listed in Table 2.3A.

The longitudinal association between FM-UE and EmNSA-UE was no longer significant after adjusting for progress of time.



**Figure 2.2** | Time course of FM-UE and EmNSA-UE over the first 26 weeks post-stroke. A FM-UE score <18 points was considered a low baseline score; >18 points a high baseline score. An EmNSA-UE score <9 points was considered a low baseline score; >9 points a high baseline score. Abbreviations: Erasmus modification of the Nottingham sensory assessment of the upper extremity (EmNSA-UE), Fugl-Meyer motor assessment of the upper extremity (FM-UE), baseline assessment within 3 weeks post-stroke (B), week 5 measurement (W5), week 12 measurement (W12), week 26 measurement (W26).

Table 2.2 | Association model of EmNSA-UE and time

A	Descriptives per measurement					
EmNSA-UE	Baseline	W5	W12	W26		
Med	24	36	39	39.5		
Iqr	[2-34]	[8-39]	[23.5-40]	[24.5-40]		
FM-UE						
Med	7	12	20	24		
Iqr	[4-30]	[5-51]	[8-58]	[7.75-57]		
Association model of EmNSA-UE and time						
EmNSA-UE	Baseline	Base to W5	Base to W12	Base to W26	W5 to W12	W5 to W26
$\beta$	19.0	7.8	11.2	12.5	3.5	4.8
CI	[16.1-21.9]	[6.0-9.6]	[9.5-13.1]	[10.8-14.3]	[1.6-5.3]	[2.9-6.6]
P	-	<0.01	<0.01	<0.01	<0.01	<0.01
						W12 to W26
						1.3
						[-0.5-3.1]
						0.15

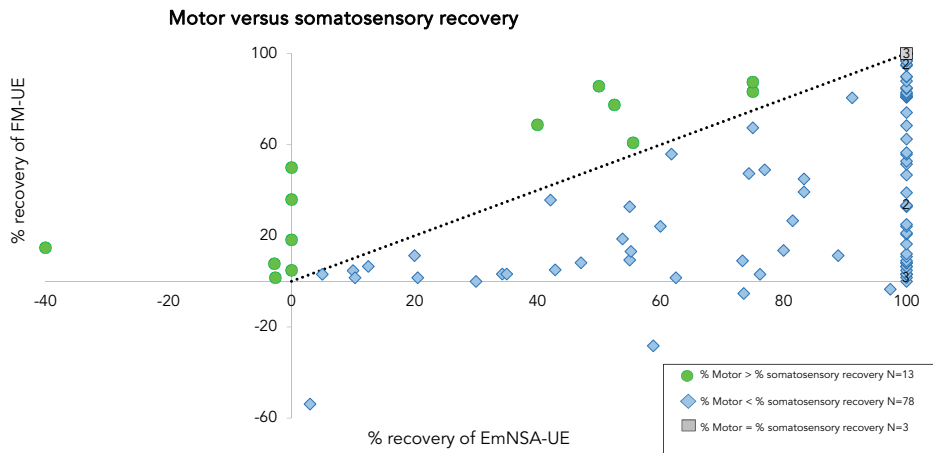
Abbreviations: Erasmus modification of the Nottingham sensory assessment of the upper extremity (EmNSA-UE), baseline (base) i.e. first measurement of each subject within 3 weeks post-stroke, week of measurement (w), beta estimate ( $\beta$ ), median (med), interquartile range (IQR), 95% confidence interval (CI), probability value for the tested model (P).

### Categorized baseline level of somatosensory impairment

The ROC-curve for somatosensory impairment showed an optimal cut-off point at 9 points on the EmNSA-UE. Of the 95 patients with a somatosensory impairment at baseline, 37 (39.0%) had an EmNSA-UE score lower than 9 points and were categorized as having a severe somatosensory impairment. Eight patients had a low baseline score on the EmNSA-UE while having a high baseline score on the FM-UE. Thirty-two patients had a high baseline score on the EmNSA-UE while having a low baseline score on the FM-UE. The remaining 54 patients showed either a high (26 patients) or a low (28 patients) baseline scores for both modalities.

### Influence of severe baseline somatosensory impairment on motor recovery

The longitudinal association between motor and somatosensory impairments did not differ significantly between patients with a high or a low level of somatosensory impairment at baseline, as no significant interaction effect was found between longitudinal EmNSA-UE score and baseline EmNSA-UE level,  $P=0.09$ .



**Figure 2.3** | Percentage of motor and somatosensory recovery between baseline (within 3 weeks post-stroke) and 26 weeks post-stroke.

Motor and somatosensory recovery of 94 patients expressed as a percentage of the maximum possible improvement: (EmNSA-UE recovery=EmNSA-UE-26weeks/ (40 – EmNSA-UE-baseline)\*100%), (FM-UE recovery=FM-UE-26weeks/ (66 – FM-UE-baseline)\*100%).

The black dashed line represents the same percentage recovery of both modalities. When patients show relatively more somatosensory than motor recovery, their value (blue diamond) is below the dashed line,  $N=78$ . When patients show relatively more motor than somatosensory recovery, their value (green dot) is above the dashed line,  $N=13$ . Three patients showed 100% recovery of both modalities (gray square in top corner). Abbreviations: Erasmus modification of the Nottingham sensory assessment of the upper extremity (EmNSA-UE), Fugl-Meyer motor assessment of the upper extremity (FM-UE).

Table 2.3A | Association models of FM-UE and EmNSA-UE

		Association model of FM-UE and EmNSA-UE									
FM-UE	Constant	EmNSA									
$\beta$	10.91	0.55									
CI	[5.78–16.04]	[0.43–0.67]									
P	-	<0.01									
		Association model of FM-UE and EmNSA-UE adjusted for covariates									
FM-UE	Constant	EmNSA	Age	Lesion side right	CIRS	NIHSS -adapt	LCT	MI-LE			
$\beta$	3.42	0.21	0.02	-3.44	1.90	-2.20	-0.31	0.34			
CI	[-13.38–20.21]	[0.06–0.35]	[-0.22–0.26]	[-10.49–3.61]	[0.51–3.29]	[-3.81–0.60]	[-0.62–0.00]	[0.26–0.42]			
P	-	<0.01	0.86	0.33	<0.01	<0.01	0.05	<0.01			
		Association model of FM-UE and EmNSA-UE adjusted for covariates and time									
FM-UE	Constant	EmNSA	Age	Lesion side right	CIRS	NIHSS -adapt	LCT	MI-LE	Base to W5	Base to W12	Base to W26
$\beta$	9.81	0.13	-0.02	-4.11	2.15	-1.02	-0.45	0.21	5.60	7.83	9.06
CI	[-10.41–30.03]	[-0.03–0.28]	[-0.31–0.28]	[-12.81–4.60]	[0.40–3.90]	[-2.71–0.68]	[-0.76–0.15]	[0.12–0.30]	[2.01–9.19]	[3.72–11.95]	[4.83–13.29]
P	-	0.10	0.91	0.35	0.02	0.24	<0.01	<0.01	<0.01	<0.01	<0.01

Table 2.3B | Association models of FM-UE and EmNSA-UE, with interaction effects of baseline FM-UE level and recovery pattern groups

B Association model of FM-UE and EmNSA-UE, for low/high FM-UE score at baseline, adjusted for covariates and time																	
FM-UE	β	CI	P	EmNSA low FM-UE base		Constant high FM-UE base		EmNSA high FM-UE base		Lesion side right	CIRS	NIHSS -adapt	LCT	MI-LE	Base to W5	Base to W12	Base to W26
				β	CI	β	CI	β	CI								
	2.75	[-10.11–15.61]		0.05	[-0.09–0.19]	8.03–37.9%	[8.03–37.9%	0.44	[0.22–0.65]	-0.10–0.26]	0.08	-2.35–0.52]	-0.35	0.15	5.9	8.45	9.7
	-			0.46	[-0.09–0.19]	-		<0.01		0.06	0.89	0.21	0.01	<0.01	<0.01	<0.01	<0.01
Association model of FM-UE and EmNSA-UE in N=60 with a low baseline FM-UE score, for recoverers/non-recoverers, adjusted for covariates and time																	
FM-UE	β	CI	P	EmNSA rec		Constant non-rec		EmNSA non-rec		Lesion side right	CIRS	NIHSS -adapt	LCT	MI-LE	Base to W5	Base to W12	Base to W26
				β	CI	β	CI	β	CI								
	3.97	[-12.33–20.28]		0.69	[0.4–0.90]	-2.74–25.31]		11.29	[-0.30–0.03]	-0.03	0.18	-1.51	-0.26	0.07	4.85	7.71	9.25
	-			<0.01				0.02		0.76	0.77	0.04	0.05	0.15	0.01	<0.01	<0.01

Abbreviations: Fugl-Meyer motor assessment of the upper extremity (FM-UE), Erasmus modification of the Nottingham sensory assessment of the upper extremity (EmNSA-UE), cumulative illness rating score (CIRS), national institute of stroke scale without items 5a, 5b, 6a, 6b, 7, 8 and 11 (NIHSS-adapt), letter cancellation task (LCT), motricity index for lower extremity (MI-LE), baseline (base) i.e. first measurement of each subject within 3 weeks post-stroke, baseline FM-UE score below 18 (low base), baseline FM-UE score 18 or higher (high base), recoverers (rec), i.e. FM-UE 18 or higher at 26 weeks post-stroke and at least a 6-point improvement between baseline and 26 weeks post-stroke, non-recoverers (non-rec), i.e. FM-UE below 18 at 26 weeks post-stroke or failed to show 6-point improvement between baseline and 26 weeks post-stroke; week of measurement (w), beta estimate (β), 95% confidence interval (CI), probability value for the tested model (P).



***Categorized baseline level of motor impairment: recoverers and non-recoverers***

The ROC-curve for motor impairment confirmed the optimal cut-off point of 18 points on the FM-UE. Of the 195 patients with a motor impairment at baseline, 109 (55.9%) had an FM-UE score lower than 18 points, and were categorized as having a severe motor impairment of which 60 were included in the analyses due to a somatosensory impairment. Of these 60 patients with a low baseline score, 21 were categorized as recoverers and 39 as non-recoverers, based on their FM-UE score at 26 weeks post-stroke.

***Influence of somatosensory recovery on motor recovery, high versus low baseline and recoverers versus non-recoverers***

A significant interaction effect was found between longitudinal EmNSA-UE and baseline FM-UE levels. For the group with a high baseline score (FM-UE $\geq$ 18) this resulted in a significant longitudinal association between FM-UE and EmNSA-UE:  $\beta$  intercept=2.75,  $\beta$  EmNSA-UE=0.44,  $P<0.01$ . Among the group with a low FM-UE baseline score, no significant association was found between longitudinal FM-UE and EmNSA-UE scores.

A significant interaction effect was also found between longitudinal EmNSA-UE and motor recovery pattern, with a significant positive association for the recoverers:  $\beta$  intercept=3.97,  $\beta$  EmNSA-UE=0.69,  $P<0.01$ , while the non-recoverers showed a negative association:  $\beta$  intercept=11.29,  $\beta$  EmNSA-UE=-0.16,  $P=0.02$ . Table 2.3B gives all corresponding values of the association model between motor and somatosensory impairments for the groups based on baseline level of motor impairment and motor recovery pattern.

***Influence of somatosensory recovery on motor recovery, based on between- and within-subject effects***

Table 2.4 shows the individual between- and within-subject effects of the association between motor and somatosensory impairments.

Both the between- and within-subject parts showed a significant longitudinal association between FM-UE and EmNSA-UE:  $\beta$  intercept=10.22,  $\beta$  EmNSA-UE/between=0.57,  $P<0.01$  and  $\beta$  EmNSA-UE/within=0.54,  $P<0.01$ .

When correcting for age, lesion side, LCT, CIRS, NIHSS-adapt, and MI-LE, only the between-subject association between FM-UE and EmNSA-UE remained significant:  $\beta$  intercept=-1.15,  $\beta$  EmNSA-UE/between=0.35,  $P<0.01$  and  $\beta$  EmNSA-UE/within=0.12,  $P=0.16$ . The between association increased after adjustment for progress of time, to:  $\beta$  intercept=-0.21,  $\beta$  EmNSA-UE/between=0.49,  $P<0.01$  and  $\beta$  EmNSA-UE/within=0.00,  $P=0.98$ .

Table 2.4 | Hybrid association models of FM-UE and EmNSA-UE

Hybrid association model of FM-UE and EmNSA-UE											
FM-UE	EmNSA /between	EmNSA /within									
Constant											
$\beta$	0.57	0.54									
CI	[0.27–0.88]	[0.41–0.68]									
P	<0.01	<0.01									
Hybrid association model of FM-UE and EmNSA-UE adjusted for covariates											
FM-UE	EmNSA /between	EmNSA /within	Age	Lesion side right	CIRS	NIHSS -adapt	LCT	MI-LE			
Constant											
$\beta$	0.35	0.12	0.02	-2.62	1.73	-2.34	-0.28	0.35			
CI	[0.12–0.58]	[-0.05–0.30]	[-0.21–0.25]	[-9.57–4.33]	[0.36–3.10]	[-3.95–0.74]	[-0.59–0.03]	[0.27–0.43]			
P	<0.01	0.16	0.87	0.45	0.01	<0.01	0.08	<0.01			
Hybrid association model of FM-UE and EmNSA-UE adjusted for covariates and time											
FM-UE	EmNSA /between	EmNSA /within	Age	Lesion side right	CIRS	NIHSS -adapt	LCT	MI-LE	Base to W5	Base to W12	Base to W26
Constant											
$\beta$	0.49	-0.00	-0.03	-2.11	1.78	-0.88	-0.45	0.20	7.03	9.87	11.29
CI	[0.20–0.77]	[-0.18–0.18]	[-0.32–0.26]	[-10.71–6.48]	[0.06–3.51]	[-2.56–0.81]	[-0.75–0.15]	[0.11–0.29]	[3.34–10.73]	[5.56–14.19]	[6.83–15.76]
P	<0.01	0.98	0.83	0.62	0.04	0.31	<0.01	<0.01	<0.01	<0.01	<0.01

Abbreviations: Fugl-Meyer motor assessment of the upper extremity (FM-UE), Erasmus modification of the Nottingham sensory assessment of the upper extremity (EmNSA-UE), cumulative illness rating score (CIRS), national institute of stroke scale without items 5a, 5b, 6a, 6b, 7, 8 and 11 (NIHSS-adapt), letter cancellation task (LCT), motricity index for lower extremity (MI-LE), baseline (base) i.e. first measurement of each subject within 3 weeks post-stroke, week of measurement (w), beta estimate ( $\beta$ ), 95% confidence interval (CI), probability value for the tested model (P).

No significant interaction effects were found between the baseline EmNSA-UE and FM-UE levels, or the baseline EmNSA-UE level and the pattern of spontaneous motor recovery.

## DISCUSSION

In the present study, we investigated the longitudinal association between motor and somatosensory impairments, in a cohort of 94 patients with a first-ever ischemic stroke, measured at four fixed time points during the first 6 months post-stroke.

We show that motor recovery was significantly longitudinally associated with somatosensory recovery. Both modalities recover within the same time window of spontaneous neurobiological recovery in the first 3 months post-stroke. After adjusting for possible confounders such as age and co-morbidities, the association between motor and somatosensory impairment remained significant, underpinning its robustness. However, the association disappeared when correcting for progress of time, suggesting that time-dependent change due to spontaneous neurobiological recovery is the main factor that drives improvement of both modalities in the same time window<sup>49,50</sup>. In the longitudinal association model between FM-UE and EmNSA-UE, we found no significant influence of baseline level of somatosensory impairment. The current findings therefore suggest that severe somatosensory impairment in the first weeks post-stroke does not necessarily obstruct motor recovery.

However, we did find evidence that somatosensory function is an important factor to achieve full recovery of motor impairment. None of the patients with full motor recovery showed impaired somatosensory recovery. Our results suggest that different mechanisms are relevant in subgroups of patients who show or do not show spontaneous neurobiological motor recovery of the upper paretic limb. In patients with a low level of motor impairment at baseline (FM-UE >18) and/or significant spontaneous improvement of motor impairment over time (i.e. recoverers), an association between motor and somatosensory impairments was present, irrespective of progress of time. This finding suggests that motor recovery is influenced, in a direct manner, by the recovery of somatosensory impairment.

Meyer et al.<sup>51</sup> studied somatosensory recovery in 32 patients after stroke in the first week and at 26 weeks post-stroke. They found only very low correlations between the FM-UE and the subdomains of the EmNSA-UE in the first week post-stroke ( $r=-0.03$  –  $-0.14$ ), while low to moderate correlations were found at 26 weeks post-stroke ( $r=0.02$ – $0.27$ )<sup>51</sup>. We found, in line with these results that severe baseline somatosensory impairment does not necessarily prevent spontaneous motor recovery as hypothesized, but rather that recovery of somatosensory impairment is required for achieving full motor recovery.

## Differences in mechanisms, based on between- and within-subject effects

The hybrid association model showed that the within-subject effect, which represents the progress of time after stroke and thus spontaneous neurobiological recovery, is influenced by factors such as stroke severity. However, we found a clear between-subject effect in the association between motor and somatosensory impairments, which remained significant after correcting for covariates and progress of time. This result reflects the same general concept as was captured in the analyses using motor recovery pattern subgroups based on cut off grouping variables. Somatosensory impairment affects motor recovery, which supports an underlying mechanism consistent with processes of learning-dependent plasticity.

While the hybrid model does not give insight into the existence of subgroups of recoverers and non-recoverers, findings do confirm that the association between motor and somatosensory impairments varies between patients with different motor recovery patterns. Note that using a hybrid model may circumvent inherent problems of defining cut off values in small groups of patients, and may therefore be recommended as an instrument to separate the within-subject variance from the between-subject variance in explaining neurobiological recovery in repeated measurement designs <sup>48</sup>.

## Limitations

The EmNSA-UE was used to measure somatosensory impairment, as has been recommended due to its good to excellent reliability <sup>2,37</sup>. The standardized response mean of the revised NSA has a wide range, from 0.34 to 0.83, depending on the subdomain <sup>52</sup>. The smallest detectable change or minimal clinically relevant difference of the EmNSA has however not been determined. The EmNSA-UE is a broad measure focusing on detection of impairments in the primary somatosensory modalities using a subjective ordinal 3-point scale and does not evaluate somatosensory discrimination, such as tactile 2-point discrimination. It can be hard to obtain an accurate and valid score for patients with cognitive or attention impairments. Hence, we assumed a liberal measurement error of at least 2 points when defining somatosensory impairments.

While multiple relevant covariates have been taken into account in this study, we could not correct for lesion volume or location in our association model, since this information was not available. The results from our study however suggest that the association between the recovery of motor and somatosensory impairments is based on more than the close anatomical distance of somato-motor brain areas, the overlap of the lesion in metabolism-dependent systems and the recovery of penumbral tissue <sup>53,54</sup>. Type of treatment was also

not explicitly accounted for as a potential covariate in the present study, although there is no evidence for a confounding effect <sup>32</sup>.

### Future directions

Although the underlying neurophysiological mechanisms for motor and somatosensory impairments could not be causally linked in the current study, our results do provide direction for future research.

As has been shown in animal models, reduced somatosensory input compromises learning-dependent plasticity <sup>28-31</sup>. The absence of recovery of somatosensory impairment could potentially compromise learning-dependent plasticity after stroke, resulting in inferior motor recovery. Recovery of sensorimotor impairment after stroke is associated with changes in resting-state functional connectivity (RS-FC) in humans and rodents <sup>55,56</sup>. Hakon et al. recently showed, in an animal model of stroke, that multisensory stimulation through exposure to an enriched environment improves tactile-proprioceptive function and RS-FC in mice, as compared to those housed in a standard environment <sup>57</sup>. Clinically, the present findings suggest that somatosensory retraining could be beneficial in particular for those patients who show incomplete motor recovery post-stroke, as was suggested in a recent systematic review of Turville et al. <sup>58</sup>. The evidence regarding the effectiveness of available somatosensory retraining programs on sensorimotor function is currently however limited <sup>58-60</sup>. In this vein, it is important to highlight that several items on the FM-UE, like the pincer, spherical and cylindrical grasp can be considered tasks that depend on sensorimotor function <sup>12</sup> and that achieving a full FM-UE score will depend on optimal sensorimotor function. Animal models in which the somatosensory and visual impairments are selectively lesioned may give new insights into whether rehabilitation interventions might be able to interact with motor recovery via learning-dependent plasticity <sup>9,24</sup>.

Clinical practice would highly benefit from measures that can more objectively establish somatosensory function. Therefore, one may consider neuroimaging <sup>61</sup> and specifically diffusion tensor imaging to determine the intactness of structural pathways after stroke <sup>18</sup>. Neurophysiological techniques that target the intactness of the somatosensory system such as, closed-loop identification techniques by applying position perturbations <sup>62-64</sup> or somatosensory and median nerve stimulation to the affected arm <sup>65</sup>, may be more precise ways to test the integrity of somatosensory pathways after stroke than clinical measures <sup>63-65</sup>. However, the added prognostic value of these non-invasive techniques above clinical testing alone needs further investigation within the first days post-stroke.

Imaging parameters of the lesion are needed in future studies to corroborate our current results, which are based on clinical measures of impairment. One of the few imaging studies investigating somatosensory impairment reported a greater lesion load in the corticospinal tracts of patients (N=32) with impaired ability to perceive a somatosensory stimulus (e.g. touch, pressure) at 4–7 days post-stroke, yet all of the patients showed full recovery of this somatosensory modality at 26 weeks post-stroke<sup>18</sup>. The authors did, however, not study the relationship with motor recovery. Longitudinal imaging studies relating functional connectivity patterns to motor and somatosensory impairment and recovery are needed to provide more insights into connectional diaschisis and network changes in post-stroke recovery<sup>66</sup>.

Recently, Hope et al. highlighted that the 70% proportional recovery rule maybe mathematically inflated<sup>67</sup>. The proportional recovery model is also vulnerable for ceiling effects and may therefore give a too optimistic impression of the predictability of outcomes<sup>67</sup>. Prognostic mixture models, not suffering from mathematical inflation, may be a next step to improve early individual clinical decision making at stroke units<sup>68</sup>. Beyond this discussion<sup>69</sup>, our results indicate that somatosensory recovery is important to explain variability in the percentage of motor recovery, specifically in the subgroup of recoverers. Our results are a first step towards pinpointing factors that may interfere with, and/or prevent spontaneous motor recovery in patients early after stroke. Understanding of these factors, such as somatosensory impairment, is needed to develop strategies to optimize quality of movement after stroke<sup>70</sup>.

## Acknowledgements

We would like to thank prof. dr. Jos. W. Twisk for his help with the statistical analysis in this chapter.

## Funding

This study was financially supported by a fellowship grant from the Dutch brain foundation, the Netherlands (Hersenstichting, F2011(1)-25), a European advanced grant (ERC-AG-ID1, FP7-IDEAS-ERC, 291339) and a ZonMw grant (89000001) from the Dutch Organization for Health Research and Development.



## REFERENCES

- 1 Tyson SF, Hanley M, Chillala J, Selley AB, Tallis RC. Sensory loss in hospital-admitted people with stroke: characteristics, associated factors, and relationship with function. *Neurorehabil Neural Repair* 2008; **22**: 166–172.
- 2 Connell LA, Tyson SF. Measures of sensation in neurological conditions: a systematic review. *Clin Rehabil* 2012; **26**: 68–80.
- 3 Carey LM, Matyas TA, Baum C. Effects of somatosensory impairment on participation after stroke. *Am J Occup Ther* 2018; **72**: 1–10.
- 4 Meyer S, De Bruyn N, Lafosse C, Van Dijk M, Michielsen M, Thijs L et al. Somatosensory impairments in the upper limb poststroke: distribution and association with motor function and visuospatial neglect. *Neurorehabil Neural Repair* 2016; **30**: 731–742.
- 5 Meyer S, Karttunen AH, Thijs V, Feys H, Verheyden G. How do somatosensory deficits in the arm and hand relate to upper limb impairment, activity, and participation problems after stroke? A systematic review. *Phys Ther* 2014; **94**: 1220–1232.
- 6 Sommerfeld DK, von Arbin MH. The impact of somatosensory function on activity performance and length of hospital stay in geriatric patients with stroke. *Clin Rehabil* 2004; **18**: 149–155.
- 7 Kwakkel G, Kollen B, Twisk J. Impact of time on improvement of outcome after stroke. *Stroke* 2006; **37**: 2348–2353.
- 8 Bernhardt J, Hayward KS, Kwakkel G, Ward NS, Wolf SL, Borschmann K et al. Agreed definitions and a shared vision for new standards in stroke recovery research: The Stroke Recovery and Rehabilitation Roundtable taskforce. *Int J Stroke* 2017; **12**: 444–450.
- 9 Winters C, Kwakkel G, Van Wegen EEH, Nijland RHM, Veerbeek JM, Meskers CGM. Moving stroke rehabilitation forward: the need to change research. *NeuroRehabilitation* 2018; **43**: 19–30.
- 10 Kwakkel G, Kollen B, Lindeman E. Understanding the pattern of functional recovery after stroke: facts and theories. *Restor Neurol Neurosci* 2004; **22**: 281–299.
- 11 Bolognini N, Russo C, Edwards DJ. The sensory side of post-stroke motor rehabilitation. *Restor Neurol Neurosci* 2016; **34**: 571–586.
- 12 Blennerhassett JM, Matyas TA, Carey LM. Impaired discrimination of surface friction contributes to pinch grip deficit after stroke. *Neurorehabil Neural Repair* 2007; **21**: 263–272.
- 13 Prabhakaran S, Zarahn E, Riley C, Speizer A, Chong JY, Lazar RM et al. Inter-individual variability in the capacity for motor recovery after ischemic stroke. *Neurorehabil Neural Repair* 2008; **22**: 64–71.
- 14 Winters C, Van Wegen EEH, Daffertshofer A, Kwakkel G. Generalizability of the proportional recovery model for the upper extremity after an ischemic stroke. *Neurorehabil Neural Repair* 2015; **29**: 614–622.
- 15 Byblow WD, Stinear CM, Barber PA, Petoe MA, Ackerley SJ. Proportional recovery after stroke depends on corticomotor integrity. *Ann Neurol* 2015; **78**: 848–859.
- 16 Veerbeek, Winters C, Van Wegen EEH, Kwakkel G. Is the proportional recovery rule applicable to the lower limb after a first-ever ischemic stroke? *PLoS One* 2018; **13**: e0189279.
- 17 Smith MC, Byblow Winston D, Alan BP, Cathy SM. Proportional recovery from lower limb motor impairment after stroke. *Stroke* 2017; **48**: 1400–1403.
- 18 Boccuni L, Meyer S, Kessner SS, De Bruyn N, Essers B, Cheng B et al. Is there full or proportional somatosensory recovery in the upper limb after stroke? Investigating behavioral outcome and neural correlates. *Neurorehabil Neural Repair* 2018; **32**: 691–700.
- 19 Turville ML, Matyas TA, Blennerhassett JM, Carey LM. Initial severity of somatosensory impairment influences response to upper limb sensory retraining post-stroke. *NeuroRehabilitation* 2018; **43**: 413–423.
- 20 Lazar RM, Minzer B, Antonietto D, Festa JR, Krakauer JW, Marshall RS. Improvement in aphasia scores after stroke is well predicted by initial severity. *Stroke* 2010; **41**: 1485–1488.
- 21 Marchi NA, Ptak R, Di Pietro M, Schnider A, Guggisberg AG. Principles of proportional recovery after stroke generalize to neglect and aphasia. *Eur J Neurol* 2017; **24**: 1084–1087.

- 22 Winters C, Van Wegen EEH, Daffertshofer A, Kwakkel G. Generalizability of the maximum proportional recovery rule to visuospatial neglect early poststroke. *Neurorehabil Neural Repair* 2017; **31**: 334–342.
- 23 Nijboer TCW, Kollen BJ, Kwakkel G. The impact of recovery of visuo-spatial neglect on motor recovery of the upper paretic limb after stroke. *PLoS One* 2014; **9**: e100584.
- 24 Buma F, Kwakkel G, Ramsey N. Understanding upper limb recovery after stroke. *Restor Neural Neurosci* 2013; **31**: 707–722.
- 25 Murphy TH, Corbett D. Plasticity during stroke recovery: from synapse to behaviour. *Nat Rev Neurosci* 2009; **10**: 861–872.
- 26 Boyd LA, Hayward KS, Ward NS, Stinear CM, Rosso C, Fisher RJ et al. Biomarkers of stroke recovery: consensus-based core recommendations from the stroke recovery and rehabilitation roundtable. *Int J Stroke* 2017; **12**: 480–493.
- 27 Langhorne P, Bernhardt J, Kwakkel G. Stroke rehabilitation. *Lancet* 2011; **377**: 1693–1702.
- 28 Trachtenberg JT, Chen BE, Knott GW, Feng G, Sanes JR, Welker E et al. Long-term in vivo imaging of experience-dependent synaptic plasticity in adult cortex. *Nature* 2002; **420**: 788–794.
- 29 Glencross DJ. Chapter 1 Motor control and sensory-motor integration. In: *Motor control and sensory motor integration*. North-Holland, 1995, pp 3–7.
- 30 Nudo RJ, Plautz EJ, Frost SB. Role of adaptive plasticity in recovery of function after damage to motor cortex. *Muscle Nerve* 2001; **24**: 1000–1019.
- 31 Feldman DE, Brecht M. Map plasticity in somatosensory cortex. *Science* 2005; **310**: 810–815.
- 32 Kwakkel G, Winters C, Van Wegen EEH, Nijland RHM, Van Kuijk AAA, Visser-Meily A et al. Effects of unilateral upper limb training in two distinct prognostic groups early after stroke: The EXPLICIT-Stroke randomized clinical trial. *Neurorehabil Neural Repair* 2016; **30**: 804–816.
- 33 Kwakkel G, Meskers CGM, Van Wegen EE, Lankhorst GJ, Geurts ACH, van Kuijk A et al. Impact of early applied upper limb stimulation: the EXPLICIT-stroke programme design. *BMC Neurol* 2008; **8**: 49.
- 34 Veerbeek J, Van Wegen E, Van Peppen R, Van Der Wees PJ, Hendriks E, Rietberg M et al. What is the evidence for physical therapy poststroke? A systematic review and meta-analysis. *PLoS One* 2014; **9**: e87987.
- 35 Tombaugh TN, McIntyre NJ. The Mini-Mental State Examination: A Comprehensive Review. *J Am Geriatr Soc* 1992; **40**: 922–935.
- 36 Saa JP, Tse T, Baum C, Cumming T, Josman N, Rose M et al. Longitudinal evaluation of cognition after stroke – A systematic scoping review. *PLoS One* 2019; **14**: e0221735.
- 37 Stolk-Hornsveld F, Crow JL, Hendriks EP, Van Der Baan R, Harmeling-van Der Wel BC. The Erasmus MC modifications to the (revised) Nottingham Sensory Assessment: a reliable somatosensory assessment measure for patients with intracranial disorders. *Clin Rehabil* 2006; **20**: 160–172.
- 38 Choi JC, Kim BJ, Han M-K, Lee SJ, Kang K, Park J-M et al. Utility of items of baseline national institutes of health stroke scale as predictors of functional outcomes at three months after mild ischemic stroke. *J Stroke Cerebrovasc Dis* 2017; **26**: 1306–1313.
- 39 Sanford J, Moreland J, Swanson LR, Stratford PW, Gowland C. Reliability of the Fugl-Meyer assessment for testing motor performance in patients following stroke. *Phys Ther* 1993; **73**: 447–454.
- 40 Duncan PW, Propst M, Nelson SG. Reliability of the Fugl-Meyer assessment of sensorimotor recovery following cerebrovascular accident. *Phys Ther* 1983; **63**: 1606–1610.
- 41 Gladstone DJ, Danells CJ, Black SE. The Fugl-Meyer Assessment of motor recovery after stroke: a critical review of its measurement properties. *Neurorehabil Neural Repair* 2002; **16**: 232–240.
- 42 Nakayama H, Jorgensen HS, Raaschou HO, Olsen TS. The influence of age on stroke outcome. The Copenhagen Stroke Study. *Stroke* 1994; **25**: 808–813.
- 43 Veerbeek JM, Kwakkel G, van Wegen EEH, Ket JCF, Heymans MW. Early prediction of outcome of activities of daily living after stroke: a systematic review. *Stroke* 2011; **42**: 1482–1488.
- 44 de Groot V, Beckerman H, Lankhorst GJ, Bouter LM. How to measure comorbidity. a critical review of available methods. *J Clin Epidemiol* 2003; **56**: 221–229.

- 45 Ferber S, Karnath HO. How to assess spatial neglect--line bisection or cancellation tasks? *J Clin Exp Neuropsychol* 2001; **23**: 599–607.
- 46 Collin C, Wade D. Assessing motor impairment after stroke: a pilot reliability study. *J Neurol Neurosurg Psychiatry* 1990; **53**: 576–579.
- 47 Williams LS, Yilmaz EY, Lopez-Yunez AM. Retrospective assessment of initial stroke severity with the NIH Stroke Scale. *Stroke* 2000; **31**: 858–862.
- 48 Twisk JW, de Vente W. Hybrid models were found to be very elegant to disentangle longitudinal within and between subject relationships. *J Clin Epidemiol* 2019; **107**: 66–70.
- 49 Kwakkel G, Kollen B, Twisk J. Impact of time on improvement of outcome after stroke. *Stroke* 2006; **37**: 2348–2353.
- 50 Winters C, Kwakkel G, Nijland R, Van Wegen E. When does return of voluntary finger extension occur post-stroke? A prospective cohort study. *PLoS One* 2016; **11**: e0160528.
- 51 Meyer S, De Bruyn N, Krumlinde-Sundholm L, Peeters A, Feys H, Thijs V et al. Associations between sensorimotor impairments in the upper limb at 1 week and 6 months after stroke. *J Neurol Phys Ther* 2016; **40**: 186–195.
- 52 Wu C, Chuang I, Ma H, Lin K, Chen C. Validity and responsiveness of the Revised Nottingham Sensation Assessment for outcome evaluation in stroke rehabilitation. *Am J Occup Ther* 2016; **70**: 1–8.
- 53 Heiss W-D. The ischemic penumbra: correlates in imaging and implications for treatment of ischemic stroke. The Johann Jacob Wepfer award 2011. *Cerebrovasc Dis* 2011; **32**: 307–320.
- 54 Heiss W-D, Zaro Weber O. Validation of MRI determination of the Penumbra by PET measurements in ischemic stroke. *J Nucl Med* 2017; **58**: 187–193.
- 55 van Meer MPA, van der Marel K, Wang K, Otte WM, el Bouazati S, Roeling TAP et al. Recovery of sensorimotor function after experimental stroke correlates with restoration of resting-state interhemispheric functional connectivity. *J Neurosci* 2010; **30**: 3964–3972.
- 56 Chang-hyun P, Hyuk CW, Hoon OS, Tae KS, Young BO, Alvaro P-L et al. Longitudinal changes of resting-state functional connectivity during motor recovery after stroke. *Stroke* 2011; **42**: 1357–1362.
- 57 Hakon J, Quattromani MJ, Sjolund C, Tomasevic G, Carey L, Lee J-M et al. Multisensory stimulation improves functional recovery and resting-state functional connectivity in the mouse brain after stroke. *NeuroImage Clin* 2018; **17**: 717–730.
- 58 Turville ML, Cahill LS, Matyas TA, Blennerhassett JM, Carey LM. The effectiveness of somatosensory retraining for improving sensory function in the arm following stroke: a systematic review. *Clin Rehabil* 2019; **33**: 834–846.
- 59 Doyle S, Bennett S, Fasoli SE, McKenna KT. Interventions for sensory impairment in the upper limb after stroke. *Cochrane database Syst Rev* 2010; **16**: CD006331.
- 60 Schabrun SM, Hillier S. Evidence for the retraining of sensation after stroke: a systematic review. *Clin Rehabil* 2009; **23**: 27–39.
- 61 Carey LM, Abbott DF, Harvey MR, Puce A, Seitz RJ, Donnan GA. Relationship between touch impairment and brain activation after lesions of subcortical and cortical somatosensory regions. *Neurorehabil Neural Repair* 2011; **25**: 443–457.
- 62 Meskers CGM, de Groot JH, de Vlugt E, Schouten AC. NeuroControl of movement: system identification approach for clinical benefit. *Front Integr Neurosci* 2015; **9**: 48.
- 63 Campfens SF, Zandvliet SB, Meskers CGM, Schouten AC, van Putten MJAM, van der Kooij H. Poor motor function is associated with reduced sensory processing after stroke. *Exp Brain Res* 2015; **233**.
- 64 Zandvliet SB, van Wegen EEH, Campfens SF, van der Kooij H, Kwakkel G, Meskers CGM. Position-cortical coherence as a marker of afferent pathway integrity early post-stroke, a prospective cohort study. *Neurorehabil Neural Repair* 2020; **34**: 344–359.
- 65 Kalogiannia K, Saes M, Vlaar MP, Wegen EE. van, Kwakkel G, Schouten AC et al. Are longitudinal SSEP recordings a biomarker for proportional motor recovery post stroke? *Submitted*.
- 66 Carrera E, Tononi G. Diaschisis: past, present, future. *Brain* 2014; **137**: 2408–2422.
- 67 Hope TMH, Friston K, Price CJ, Leff AP, Rotshtein P, Bowman H. Recovery after stroke: not so proportional after all? *Brain* 2019; **142**: 15–22.

- 68 van der Vliet R, Selles RW, Andrinopoulou E-R, Nijland R, Ribbers GM, Frens MA *et al.* Predicting upper limb motor impairment recovery after stroke: a mixture model. *Ann Neurol* 2020; **87**: 383–393.
- 69 Kundert R, Goldsmith J, Veerbeek JM, Krakauer JW, Luft AR. What the proportional recovery rule is (and is not): methodological and statistical considerations. *Neurorehabil Neural Repair* 2019; **33**: 875–875.
- 70 Kwakkel G, Van Wegen E, Burridge JH, Winstein CJ, Van Dokkum L, Alt Murphy M *et al.* Standardized measurement of quality of upper limb movement after stroke: Consensus-based core recommendations from the Second Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2019; **14**: 783–791.







# Chapter 3

Is resting-state EEG longitudinally associated with recovery of clinical neurological impairments early post-stroke? A prospective cohort study

Mique Saes\*

Sarah B. Zandvliet\*

Aukje S. Andringa

Andreas Daffertshofer

Jos W.R. Twisk

Carel G.M. Meskers

Erwin E.H. van Wegen

Gert Kwakkel

on behalf of the 4D-EEG consortium

\*Authors contributed equally

*Neurorehabilitation and Neural Repair* 2020;  
34(5): 389–402

## ABSTRACT

**Background:** The time course of cortical activation and its relation with clinical measures may elucidate mechanisms underlying spontaneous neurobiological recovery after stroke.

**Objective:** We aimed to investigate (1) the time course of cortical activation as revealed by EEG-based spectral characteristics during awake rest and (2) the development of these spectral characteristics in relation to global neurological and upper-limb motor recovery in the first 6 months post-stroke.

**Methods:** Resting-state EEG was measured serially in 41 patients after a first-ever ischemic stroke, within 3 weeks and at 5, 12 and 26 weeks post-stroke. We computed the brain symmetry index (BSI) and directional BSI (BSI<sub>dir</sub>) over different frequency bands (1–25 Hz, delta, theta) and delta/alpha ratio (DAR). The national institutes of health stroke scale (NIHSS) and Fugl-Meyer motor assessment of the upper extremity (FM-UE) were determined as clinical reflections of spontaneous neurobiological recovery. Longitudinal changes in spectral characteristics and within- and between-subject associations with NIHSS and FM-UE were analyzed with linear mixed models.

**Results:** Spectral characteristics showed a gradual normalization over time, within and beyond 12 weeks post-stroke. Significant within- and between-subject associations with NIHSS were found for BSI<sub>dir<sub>delta</sub></sub> and DAR of the affected hemisphere (DAR<sub>AH</sub>). BSI<sub>dir<sub>delta</sub></sub> also demonstrated significant within- and between-subject associations with FM-UE.

**Conclusions:** Changes in spectral characteristics are not restricted to the time window of recovery of clinical neurological impairments. The present study suggests that decreasing DAR<sub>AH</sub> and BSI<sub>dir<sub>delta</sub></sub> reflect improvement of global neurological impairments, whereas BSI<sub>dir<sub>delta</sub></sub> was also specifically associated with upper-limb motor recovery early post-stroke.

---



## INTRODUCTION

Most stroke survivors suffer from upper limb paresis in the acute phase after stroke <sup>1</sup>. About 70–80% of them will show some level of spontaneous neurobiological recovery (i.e. 'recoverers'), whereas 20–30% of patients do not recover at all (i.e. 'non-recoverers') <sup>2</sup>. Spontaneous motor recovery takes place predominantly within the first 3 months post-stroke, after which most patients reach a plateau <sup>3</sup>. The mechanisms that drive spontaneous neurobiological recovery are mainly the salvation of penumbral tissue <sup>4</sup> and spontaneous regenerative processes enhanced by an upregulation of growth-promoting factors, angiogenesis and resolution of diaschisis <sup>4,5</sup>.

The main improvements in terms of the national institutes of health stroke scale (NIHSS) and Fugl-Meyer motor assessment of the upper extremity (FM-UE) take place in the time window of spontaneous neurobiological recovery, which may extend up to ten weeks after stroke onset <sup>6</sup>. A return of brain function towards its normal neural state is associated with better behavioral outcomes after stroke <sup>7–9</sup>. The longitudinal association between clinical improvements and changes in cortical activation, and whether these changes occur within the time window of spontaneous neurobiological recovery, have hardly been investigated so far <sup>10,11</sup>.

Neuronal oscillations, measured with magneto- or electro-encephalography (MEG/EEG), have been suggested to serve as a measurement tool for potential biomarkers that can be used to study the association with behavioral recovery <sup>11</sup>. In particular, stroke is associated with increased low-frequency brain oscillations in the delta (1–4 Hz) and theta bands (4–8 Hz) <sup>12–14</sup>, as well as decreased alpha (8–12 Hz) activity <sup>15,16</sup>. A spectral characteristic quantifying this phenomenon is the delta/alpha ratio (DAR). Since stroke may lead to increased delta activity with or without decreased alpha activity, a ratio between these components may be more sensitive compared to the individual components for reflecting severity of neurological deficit, and normalization of the underlying neurological deficits due to spontaneous neurological recovery after stroke. DAR appears to correlate with the severity of global neurological impairments measured with the NIHSS <sup>17</sup> in the acute phase (<1 week) post-stroke. However, in a recent study performed in the chronic post-stroke phase (>6 months), we could not find significant differences in DAR between patients and age- and gender-matched healthy individuals, nor did we find a significant association between DAR and motor impairment as measured with the FM-UE <sup>14</sup>. The above results suggest a decrease in DAR over time across stroke patients towards normal values, regardless of global neurological impairment or motor impairment.

The pairwise derived brain symmetry index (BSI) captures brain activity lateralization, and seems to be associated with stroke severity<sup>13,17,18</sup>. Several studies have shown that BSI is increased in the early sub-acute phase (between 1 week and 3 months)<sup>17,19</sup> and in the chronic phase<sup>14</sup> post-stroke, when compared to healthy individuals. The extended directional version of the BSI showed that increased low-frequency power in the affected hemisphere relative to the unaffected hemisphere (i.e., asymmetry towards the affected side), is highly associated with decreased motor function of the upper extremity in patients with chronic stroke<sup>14</sup>. We argue that directional asymmetry measures based on low-frequency oscillations can be useful in the assessment of the asymmetry of hemispheric activity early post-stroke, whose normalization is associated with neurological recovery.

In the present observational cohort study with repeated measurements performed at fixed times post-stroke, we investigated the time course of EEG-based spectral characteristics during awake rest as a representation of neuronal deficits. We simultaneously measured the time course of global neurological recovery and upper limb motor function early post-stroke, enabling us to investigate the longitudinal associations.

We addressed the following research questions:

1. What is the time course of the spectral characteristics DAR, BSI and BSIdir within the first 6 months post-stroke?
2. Are DAR, BSI and BSIdir longitudinally associated with clinically observed improvements of the NIHSS and FM-UE?

As regards (1), we hypothesized that the spectral characteristics would change in the direction of values seen in healthy individuals<sup>14</sup>. These changes might be caused by decreasing delta activity in the affected hemisphere, and hence might be mainly reflected by the  $DAR_{AH'}$  and the BSI and BSIdir when estimated over the delta band. In addition, we hypothesized that changes would occur within the time window of spontaneous neurobiological recovery (i.e. 3 months post-stroke).

We previously found a significant association for FM-UE with BSI and BSIdir but not for DAR in the chronic phase post-stroke<sup>14</sup>. Regarding the NIHSS, literature showed a significant association in the acute phase with BSI and DAR<sup>17</sup>. Therefore, as regards (2), we hypothesized that recovery of global neurological impairment as measured with NIHSS would be positively associated with a gradual decrement (i.e. normalization) in DAR. In addition, we hypothesized that a decrease in BSI (i.e. normalization) would be associated with improvement of NIHSS and FM-UE scores within the first 3 months post-stroke.

## METHODS

### Participants

In our multicenter longitudinal cohort study, patients admitted to the stroke units of six participating hospitals from June 2015 until June 2017 were eligible for participation. Fifty-five patients were included within 3 weeks post-stroke. The inclusion criteria were: (1) first-ever ischemic stroke according to CT or MRI scan; (2) <3 weeks post-stroke; (3) upper limb paresis (NIHSS 5a/b >0); (4) ≥18 years of age; and (5) providing written informed consent. Exclusion criteria were: (1) upper extremity orthopedic limitations present prior to stroke onset; (2) recurrent stroke; and (3) severe cognitive problems, i.e. mini mental state examination score <18<sup>20</sup>. The present study (registered at the Dutch trial register as NTR4221) was approved by the medical ethics committee of the VU university medical centre, Amsterdam, the Netherlands (NL 47079 029 14) and carried out in accordance with the Code of Ethics of the world medical association (declaration of Helsinki, 2013)<sup>21</sup>.

### Procedures

High-density EEG measurements and clinical assessments were performed within the first 3 weeks and at weeks 5, 12 and 26 post-stroke. The first measurement was conducted as soon as feasible. To optimize the feasibility of assessing early sub-acute patients at fixed times post-stroke, a specially equipped van (Figure 3.1) was used to perform clinical and EEG measurements, irrespective of the patient's place of residence, such as a hospital, rehabilitation center, nursing home or their own home. With that, the burden of traveling for the patients was reduced. The measurement van was customized to allow EEG acquisition of the same quality as in our hospital setting<sup>22</sup>. The resting-state EEG measurement analyzed in the current study was part of a larger study protocol. The duration of the full EEG protocol was dependent on patient's ability to perform tasks. Including preparation of the patient this took between 45 minutes, in case only resting-state EEG was measured, and 2 hours, in case all tasks were performed.

### Electroencephalography

During the EEG measurement, patients were seated in a wheelchair and were asked to focus their eyes on a dot displayed on a flat screen. Five consecutive trials of one-minute resting-state EEG data were collected. High-density 62-channel EEG was recorded using an actively shielded EEG cap with electrode placement according to the international 10–20 system (Ag/AgCl electrodes and REFA multichannel amplifier, TMSi, Oldenzaal, the Netherlands, with ASA acquisition software, ANT software BV, the Netherlands).



Figure 3.1 | Measurement set-up in a specially equipped van.

Electrode impedances were kept below 20 k $\Omega$ . EEG signals were online referenced to average. In addition, bipolar Ag/AgCl electrodes served to monitor the muscle activity of the m. extensor carpi radialis and m. flexor carpi radialis of both arms. All signals were sampled at a rate of 2048 Hz.

### Clinical assessments

Clinical assessments encompassed the NIHSS [0–42] and FM-UE [0–66]. NIHSS is a measure of the severity of global neurological impairment to classify stroke severity<sup>23</sup>. FM-UE measures the synergy-dependent motor recovery of the upper limb. Both are

recommended as outcome measure in stroke research<sup>23-25</sup>, and the time window of their change is assumed to reflect the period of spontaneous neurobiological recovery.

## Data analysis

### *Pre-processing*

Offline analysis was conducted using Matlab (R2012a, The Mathworks, Natwick, MA) with the FieldTrip toolbox for EEG/MEG analysis<sup>26</sup>. EEG data were filtered with a 4<sup>th</sup>-order bi-directional high-pass Butterworth filter (cut-off at 0.5 Hz). Power-line artifacts were reduced using notch filters around 50, 100, and 150 Hz (4<sup>th</sup>-order bi-directional Butterworth, band-width 1 Hz). Channels without data or very poor data quality were interpolated as the weighted average of the surrounding electrodes, followed by re-referencing to the remaining average. For each measurement, an average of 0.17 electrodes were interpolated. Further artifact removal consisted of the exclusion of eye-blinks and muscle activity using independent component analysis based on visual inspection of the components' waveforms, power spectrum and topographic distributions. For each measurement, an average of 2.9 components were removed. The resulting signals were again visually inspected and segments of the data which showed remaining artifacts were removed. Analyzed epochs were as large as possible, with a maximum of one minute. Modified periodograms with a Hanning window with size equal to the epoch length served as proxies of the spectral power density per channel.

### *Spectral characteristics*

#### Delta/alpha ratio

DAR was defined as the ratio of the delta power to the alpha power. For every channel  $c$  the power of the delta and alpha frequency bands ( $f = 1, \dots, 4$  Hz and  $8, \dots, 12$  Hz, respectively) was determined as the mean of the spectral power  $P_c(f)$  over this range. The delta/alpha ratio was computed as

$$\text{DAR}_c = \frac{\langle P_c(f) \rangle_{f=1, \dots, 4 \text{ Hz}}}{\langle P_c(f) \rangle_{f=8, \dots, 12 \text{ Hz}}} \quad (1)$$

The ratios were averaged over all  $N$  EEG channels yielding the global DAR as:

$$\text{DAR} = \frac{1}{N} \sum_{c=1}^N \text{DAR}_c \quad (2)$$

In addition to the assessment over all available channels, the DAR was also calculated over the affected ( $DAR_{AH}$ ) and unaffected hemisphere ( $DAR_{UH}$ ), in which the electrodes covering the midline were not included.

### Brain symmetry index

The BSI was defined as the absolute pairwise normalized difference in spectral power between the homologous channels  $c_L$  and  $c_R$  for left and right, respectively. The difference was averaged over a range from 1 to 25 Hz (adapted from <sup>13,17</sup>) according to:

$$BSI_{cp} = \left\langle \left| \frac{P_{c_R}(f) - P_{c_L}(f)}{P_{c_R}(f) + P_{c_L}(f)} \right| \right\rangle_{f=1, \dots, 25 \text{ Hz}} \quad (3)$$

These values were averaged over all channel pairs  $cp$ :

$$BSI = \frac{2}{N} \sum_{cp=1}^{N/2} BSI_{cp} \quad (4)$$

BSI values range from zero to one, indicating maximal symmetry and asymmetry, respectively. In our earlier cross-sectional study performed in the chronic phase post-stroke ( $N=21$ ), we showed the importance of the lower frequency bands <sup>14</sup>. Therefore, next to the assessment over the 1–25 Hz range, BSI was also determined separately for the delta (1–4 Hz) and theta (4–8 Hz) frequency bands.

We supplemented the BSI by a directed version (BSI<sub>dir</sub>) to account for the direction of the asymmetry <sup>14</sup>. The computation of the BSI<sub>dir</sub> omitted the absolute value of the numerator of Eq. (3). The sign of BSI<sub>dir</sub> was chosen such that values between 0 and 1 reflected greater cortical power in the affected hemisphere compared to the unaffected hemisphere, and vice versa for values between -1 and 0.

### **Statistical analysis**

The change in spectral characteristics during the first 6 months post-stroke was investigated with linear mixed models analyses with the factor time (of measurement) as the main fixed effect. A random intercept per individual was used to correct for dependency between measurements. Separate models were used for each dependent outcome parameter (DAR, BSI, BSI<sub>dir</sub>).

The longitudinal association between spectral characteristics and clinical measures was investigated with longitudinal linear mixed model analyses using two different models.

In the first model we investigated the main effects of FM-UE and NIHSS on spectral characteristics using a linear mixed model, for each individual clinical measure. For this model we used a random intercept for each individual, whereas time was added to the model as a potential confounder and effect modifier. Second, we applied a hybrid model<sup>27</sup> for the spectral characteristics which revealed a trend or a significant longitudinal association with clinical scores measured during the first 6 months post-stroke. This model made it possible to distinguish between the between- and within-subject effects of the longitudinal relationship. The between-subject covariate was determined as the individual average value over time of the independent variable, which reveals the association regardless the development over time. The within-subject covariate was calculated as the observed value minus the individual average, which reveals whether development of the dependent and independent covariates over time within a subject are associated. Subsequently, the associations between clinical measures and spectral characteristics were analyzed, resulting in two separate regression coefficients reflecting the within- and between-subject components of the longitudinal relationship.

Statistical analyses were performed using IBM SPSS statistics for windows, version 22.0 (IBM Corp., Armonk, NY, USA). Multiple testing was accounted for using the Holm-Bonferroni method. For each model, the distribution of residuals was tested for normality by inspecting histograms and Q-Q plots.

## RESULTS

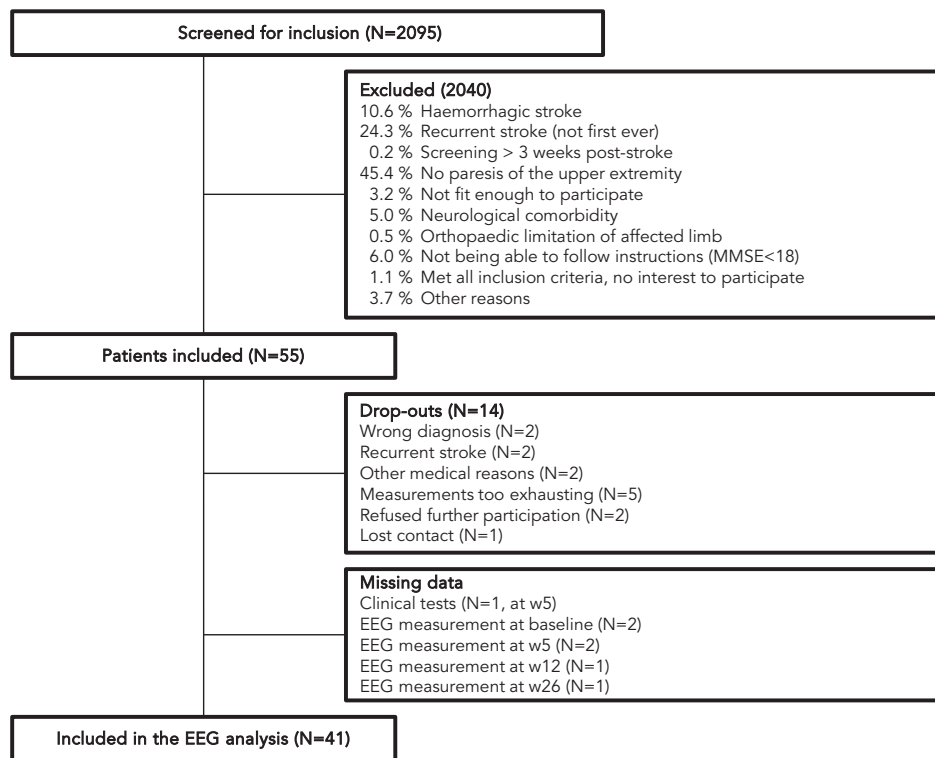
### Participants

A flowchart of the screening, inclusion and follow-up procedure, and an overview of missing data, are depicted in Figure 3.2. Forty-one out of 55 patients completed the four repeated measurements until 26 weeks post-stroke and were included in the analyses. Baseline measurements took place at  $13 \pm 5$  days (mean  $\pm$  SD) post-stroke and were repeated at w5 ( $32 \pm 3$  days), w12 ( $82 \pm 4$  days) and w26 ( $185 \pm 20$  days) post-stroke. Patient characteristics at baseline and w26 are presented in Table 3.1.

### Changes in spectral characteristics over time

Figure 3.3A-B depicts the individual and averaged time courses of the NIHSS and FM-UE scores. Visual inspection of the NIHSS and FM-UE confirms our assumption that a plateau was reached at 12 weeks post-stroke. Figure 3.3D depicts the averaged time courses of the investigated spectral characteristics. The corresponding coefficient estimates ( $\beta$ ),





**Figure 3.2** | Flowchart of screening, inclusion and follow-up.  
Abbreviations: mini mental state examination (MMSE), number of patients (N).

95% confidence intervals (CI) and probability estimates (P) are summarized in Table 3.2. Individual time courses of the spectral characteristics are presented in the supplementary materials (Figure S3.1).

DAR showed a significant decrease over time between baseline and w26 ( $\beta=-0.69$ ,  $P<0.001$ ), and from w5 to w26 ( $\beta=-0.46$ ,  $P=0.03$ ). The largest decrease was found in the affected hemisphere, while only a trend was found for the unaffected hemisphere (Figure 3.3; Table 3.2). No difference was found for BSI and  $BSI_{\theta}$  between baseline and w26, although a decrease was observed between w12 and w26 ( $\beta=-0.02$ ,  $P<0.001$ ;  $\beta=-0.02$ ,  $P<0.01$ ; Table 3.2).  $BSI_{\delta}$  showed to be decreased at w26 when compared to baseline ( $\beta=-0.02$ ,  $P=0.01$ ), w5 ( $\beta=-0.02$ ,  $P=0.01$ ) and w12 ( $\beta=-0.03$ ,  $P<0.001$ ). A statistically non-significant decrease over time was found for BSI<sub>dir</sub> and  $BSI_{dir,\theta}$  (Table 3.2), while a significant decrease was found for  $BSI_{dir,\delta}$  from baseline to w26 ( $\beta=-0.04$ ,  $P=0.003$ ), and from w5 to w26 ( $\beta=-0.03$ ,  $P=0.01$ ). This indicates that the power over the hemispheres became less lateralized especially in the lower frequency band.

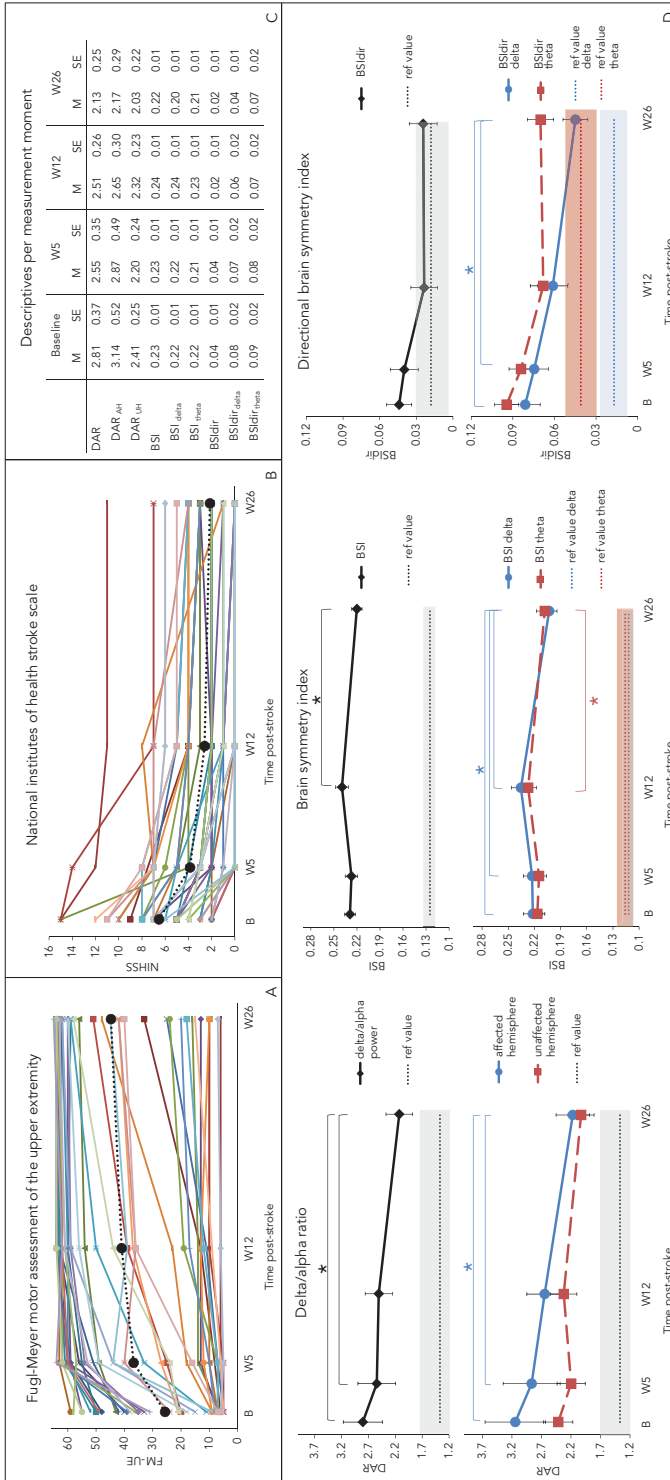
Table 3.1 | Patient demographics at baseline and 26 weeks post stroke

	All (N=41)
<b>Demographics</b>	<b>Mean (SD) or N</b>
Time PS (days)	13 (5)
Age (years)	67 (11)
Gender (male/female)	24/17
Affected hemisphere (left/right)	20/21
Bamford classification (LACI/PACI/TACI)	20/16/5
<b>Clinical scores at baseline</b>	<b>Median (IQR)</b>
NIHSS	5 (3.5–7.5)
FM-UE	21 (7–45.5)
ARAT	4 (0–32.5)
EmNSA	37 (34.5–40)
MI-UE	50 (21–72)
MI-LE	53 (32.5–75)
<b>Clinical scores at 26 weeks post-stroke</b>	<b>Median (IQR)</b>
NIHSS	2 (0–3.5)
FM-UE	58.5 (24–63)
ARAT	50 (3–57)
EmNSA	40 (38–40)
MI-UE	76 (47–84)
MI-LE	77.5 (58–100)

Demographics and clinical scores at baseline and 26 weeks post-stroke of all patients included in the analysis. Time post-stroke (time PS), i.e. time between stroke onset and baseline measurement, number of participants (N), standard deviation (SD), interquartile range (IQR), lacunar anterior circular infarct (LACI), partial anterior circulation infarct (PACI), total anterior circular infarct (TACI), national institutes of health stroke scale (NIHSS), Fugl-Meyer motor assessment of the upper extremity (FM-UE), action research arm test (ARAT), Erasmus modification of the Nottingham sensory assessment of the upper extremity (EmNSA), motricity index (MI), upper extremity (UE), lower extremity (LE).

### Association between spectral characteristics and NIHSS

Tables 3.3A–C and Table 3.4 present the longitudinal associations between spectral characteristics and NIHSS scores. A lower DAR or  $DAR_{AH}$  was longitudinally associated with a lower NIHSS score ( $\beta=0.12$ ,  $P<0.001$ ;  $\beta=0.19$ ,  $P<0.001$ ; Table 3.3A). These relations concerned significant positive within- and between-subject effects (Table 3.4). DAR and  $DAR_{AH}$  were significantly positively associated with NIHSS at baseline ( $\beta=0.12$ ,  $P=0.04$ ;  $\beta=0.21$ ,  $P=0.01$ ; Table 3.3B), while significance was not reached at other measurement moments. Regarding the  $DAR_{UH}$  no significant longitudinal association was found with NIHSS.



**Figure 3.3 |** Panel A: Development of the Fugl-Meyer motor assessment of the upper extremity over time (max=66), N=41. Panel B: Development of the national institutes of health stroke scale over time (max=42), N=41. Panel C: mean and standard error of the mean, per measurement moment, for the different spectral characteristics. Panel D: Development of the spectral characteristics over time; mean and standard errors of the mean are displayed. The blue and red dashed lines and surrounding colored area indicate the mean values of 11 healthy subjects and their standard error of the mean, as taken from Saes et al. <sup>14</sup>. \* Indicates a significant mean difference between moments, after Holm-Bonferroni corrections (P<0.05/number of comparisons). Delta band: 1–4 Hz, theta band: 4–8 Hz. Abbreviations: baseline measurement within 3 weeks post-stroke (B), measurement week post-stroke (W), delta/alpha ratio (DAR), brain symmetry index (BSI), directional BSI (BSIdir), Fugl-Meyer motor assessment of the upper extremity (FM-UE), national institutes of health stroke scale (NIHSS).

Table 3.2 | Association models of power spectral density measures and time

	Baseline to W5			Baseline to W12			Baseline to W26			W5 to W12			W5 to W26			W12 to W26		
	$\beta$	95%-CI	P	$\beta$	95%-CI	P	$\beta$	95%-CI	P	$\beta$	95%-CI	P	$\beta$	95%-CI	P	$\beta$	95%-CI	P
DAR	-0.24	(-0.65–0.18)	0.26	-0.30	(-0.72–0.12)	0.16	-0.70	(-1.11–-0.27)	<0.001	-0.06	(-0.48–0.35)	0.76	-0.46	(-0.87–-0.04)	0.03	-0.39	(-0.81–0.02)	0.06
DAR <sub>AH</sub>	-0.26	(-0.85–0.32)	0.37	-0.52	(-1.10–0.06)	0.08	-1.00	(-1.58–-0.42)	<0.001	-0.25	(-0.84–0.33)	0.39	-0.74	(-1.32–-0.15)	0.01	-0.48	(-1.06–0.09)	0.10
DAR <sub>UH</sub>	-0.19	(-0.51–0.12)	0.23	-0.08	(-0.39–0.24)	0.63	-0.39	(-0.70–-0.07)	0.02	0.11	(-0.20–0.43)	0.48	-0.19	(-0.51–0.12)	0.23	-0.31	(-0.62–0.01)	0.06
BSI	-0.00	(-0.02–0.01)	0.64	0.01	(-0.00–0.02)	0.17	-0.01	(-0.02–0.01)	0.06	0.01	(-0.00–0.02)	0.06	-0.01	(-0.02–0.003)	0.15	-0.02	(-0.03–-0.01)	<0.001
BSI <sub>delta</sub>	-0.00	(-0.02–0.02)	0.93	0.01	(-0.01–0.03)	0.22	-0.02	(-0.04–-0.01)	0.01	0.01	(-0.01–0.03)	0.19	-0.02	(-0.04–-0.00)	0.01	-0.03	(-0.05–-0.02)	<0.001
BSI <sub>theta</sub>	-0.00	(-0.02–0.01)	0.67	0.01	(-0.01–0.02)	0.27	-0.01	(-0.03–0.00)	0.12	0.01	(-0.00–0.03)	0.13	-0.01	(-0.02–0.01)	0.27	-0.02	(-0.03–-0.01)	0.008
BSIdir	-0.01	(-0.03–0.02)	0.67	-0.02	(-0.04–0.00)	0.07	-0.02	(-0.04–0.00)	0.05	-0.02	(-0.04–0.01)	0.17	-0.02	(-0.04–0.01)	0.13	-0.00	(-0.02–0.02)	0.91
BSIdir <sub>delta</sub>	-0.01	(-0.03–0.02)	0.69	-0.02	(-0.05–0.00)	0.10	-0.04	(-0.06–-0.01)	0.003	-0.02	(-0.04–0.01)	0.21	-0.03	(-0.06–-0.01)	0.01	-0.02	(-0.04–0.01)	0.17
BSIdir <sub>theta</sub>	-0.01	(-0.03–0.01)	0.42	-0.03	(-0.05–-0.00)	0.02	-0.03	(-0.05–-0.00)	0.02	-0.02	(-0.04–0.00)	0.12	-0.03	(-0.05–-0.00)	0.02	0.00	(-0.02–0.02)	0.97

Regression coefficient ( $\beta$ ), 95% confidence interval (CI), p-value (P), Holm-Bonferroni corrected significance level:  $P < (0.05/\text{number of comparisons})$ , weak (W), delta/alpha ratio (DAR), affected hemisphere (AH), unaffected hemisphere (UH), brain symmetry index (BSI), delta: mean power calculated over 1–4 Hz, theta: mean power calculated over 4–8 Hz, directional BSI (BSIdir). P-values <0.05 are shown in bold. Grey-filled boxes indicate significant values.



Table 3.3A | Association between clinical and spectral characteristics over all measurement moments corrected and uncorrected for (confounding) factor of time

	NIHSS						FM-UE					
	Uncorrected			Corrected for time			Uncorrected			Corrected for time		
	$\beta$	95% CI	P	$\beta$	95% CI	P	$\beta$	95% CI	P	$\beta$	95% CI	P
DAR	0.12	(0.05-0.19)	<0.001	0.11	(-0.001-0.21)	0.05	-0.01	(-0.03-0.001)	0.07	0.20E-2	(-0.02-0.02)	0.83
DAR <sub>AH</sub>	0.19	(0.10-0.28)	<0.001	0.18	(0.04-0.32)	0.01	-0.02	(-0.04-0.13E-2)	0.04	-12E-3	(-0.02-0.02)	0.99
DAR <sub>UH</sub>	0.05	(0.00-0.11)	0.04	0.04	(-0.04-0.12)	0.31	0.01	(-0.02-0.01)	0.32	0.29E-2	(-0.01-0.02)	0.68
BSI	0.25E-2	(0.05E-2-0.45E-2)	0.01	0.45E-2	(0.17E-2-0.74E-2)	<0.001	-0.30E-3	(-0.69E-3-0.09E-3)	0.13	-32E-3	(-0.81E-3-0.17E-3)	0.20
BSI <sub>delta</sub>	3.23E-3	(0.47E-3-5.99E-3)	0.02	0.44E-2	(0.05E-2-0.83E-2)	0.03	-0.37E-3	(-0.89E-3-0.16E-3)	0.17	-20E-3	(-0.85E-3-0.46E-3)	0.55
BSI <sub>theta</sub>	1.94E-3	(-0.41E-3-4.28E-3)	0.11	3.41E-3	(-0.03E-3-6.85E-3)	0.05	-0.27E-3	(-0.72E-3-0.19E-3)	0.25	-27E-3	(-0.84E-3-0.30E-3)	0.36
BSIdir	0.32E-2	(-0.03E-3-0.64E-2)	0.05	0.13E-2	(-0.33E-2-0.59E-2)	0.57	-0.72E-3	(-1.21E-3-0.12E-3)	0.02	-49E-3	(-1.21E-3-0.24E-3)	0.19
BSIdir <sub>delta</sub>	0.84E-2	(0.44E-2-1.23E-2)	<0.001	0.93E-2	(0.33E-2-1.53E-2)	0.003	-0.14E-2	(-0.22E-2-0.06E-2)	<0.001	-11E-2	(-0.21E-2-0.01E-2)	0.04
BSIdir <sub>theta</sub>	5.37E-3	(1.82E-3-8.93E-3)	0.003	0.39E-2	(-0.17E-2-0.95E-2)	0.17	-0.90E-3	(-1.61E-3-0.19E-3)	0.01	-46E-3	(-1.40E-3-0.49E-3)	0.34

Regression coefficient ( $\beta$ ), 95% confidence interval (CI), p-value (P), Holm-Bonferroni corrected significance level:  $P < (0.05/\text{number of comparisons})$ ; delta/alpha ratio (DAR), brain symmetry index (BSI), delta: mean power calculated over 1-4 Hz, theta: mean power calculated over 4-8 Hz, directional BSI (BSIdir), national institutes of health stroke scale (NIHSS), Fugl-Meyer motor assessment of the upper extremity (FM-UE).

**Table 3.3B** | Association between clinical and spectral characteristics per measurement moment

	Association with NIHSS per measurement moment											
	At baseline			At week 5			At week 12			At week 26		
	$\beta$	95% CI	P	$\beta$	95% CI	P	$\beta$	95% CI	P	$\beta$	95% CI	P
DAR	0.12	(0.28E-2-0.24)	0.05	0.11	(-0.02-0.24)	0.11	-0.01	(-0.17-0.15)	0.86	0.06	(-0.12-0.23)	0.52
DAR <sub>AH</sub>	0.21	(0.05-0.37)	0.01	0.17	(-0.01-0.34)	0.07	0.03	(-0.19-0.25)	0.78	0.09	(-0.15-0.33)	0.46
DAR <sub>UH</sub>	0.03	(-0.06-0.12)	0.48	0.07	(-0.03-0.17)	0.14	-0.05	(-0.17-0.07)	0.41	0.03	(-0.11-0.16)	0.70
BSI	0.48E-2	(0.14E-2-0.81E-2)	<0.01	0.36E-2	(-0.01E-2-0.72E-2)	0.06	0.63E-2	(0.18E-2-0.01)	0.006	0.52E-2	(0.03E-2-0.01E-2)	0.04
BSI <sub>delta</sub>	0.49E-2	(0.04E-2-0.94E-2)	0.03	0.34E-2	(-0.16E-2-0.84E-2)	0.18	0.57E-2	(-0.04E-2-0.01)	0.07	0.47E-2	(-0.21E-2-0.01)	0.17
BSI <sub>theta</sub>	0.34E-2	(-0.05E-2-0.74E-2)	0.09	0.25E-2	(-0.18E-2-0.68E-2)	0.26	0.61E-2	(0.07E-2-1.14E-2)	0.03	0.48E-2	(-0.10E-2-1.07E-2)	0.11
BSIdir	-0.18E-2	(-0.71E-2-0.36E-2)	0.52	0.03	(-0.43E-2-0.01)	0.25	0.698E-2	(-0.03E-2-0.01)	0.06	0.25E-2	(-0.55E-2-0.01)	0.54
BSIdir <sub>delta</sub>	0.82E-2	(0.14E-2-1.51E-2)	0.02	0.01	(0.27E-2-0.02)	0.008	0.01	(0.29E-2-0.02)	0.01	0.01	(0.33E-3-0.02)	0.04
BSIdir <sub>theta</sub>	0.27E-2	(-0.35E-2-0.89E-2)	0.39	0.47E-2	(-0.21E-2-1.15E-2)	0.18	0.76E-2	(-0.09E-2-1.60E-2)	0.08	0.01	(0.22E-2-0.02)	0.02

Regression coefficient ( $\beta$ ), 95% confidence interval (CI), p-value (P), Holm-Bonferroni corrected significance level:  $P < (0.05/\text{number of comparisons})$ , delta/alpha ratio (DAR), brain symmetry index (BSI), delta: mean power calculated over 1-4 Hz, theta: mean power calculated over 4-8 Hz, directional BSI (BSIdir), national institutes of health stroke scale (NIHSS), Fugl-Meyer motor assessment of the upper extremity (FM-UE).

Table 3.3C | Association between clinical and spectral characteristics per measurement moment

	Association with FM-UE per measurement moment											
	At baseline			At week 5			At week 12			At week 26		
	$\beta$	95% CI	P	$\beta$	95% CI	P	$\beta$	95% CI	P	$\beta$	95% CI	P
DAR	0.17E-2	(-0.02-0.03)	0.89	0.38E-2	(-0.02-0.03)	0.72	0.46E-2	(-0.02-0.03)	0.68	-0.32E-2	(-0.03-0.02)	0.77
DAR <sub>AH</sub>	-0.29E-2	(-0.04-0.03)	0.86	-0.29E-2	(-0.03-0.03)	0.79	0.17E-2	(-0.03-0.03)	0.73	-0.57E-2	(-0.04-0.02)	0.71
DAR <sub>UH</sub>	0.01	(-0.01-0.03)	0.50	0.18E-2	(-0.01-0.02)	0.83	0.58E-2	(-0.01-0.02)	0.48	-0.43E-3	(-0.02-0.02)	0.96
BSI	-0.57E-3	(-1.25 E-3-0.11 E-3)	0.10	-0.12E-3	(-0.71E-3-0.47E-3)	0.68	-0.40E-3	(-1.00E-3-0.19E-3)	0.18	-0.34E-3	(-0.94E-3-0.27E-3)	0.27
BSI <sub>delta</sub>	-0.24E-3	(-1.17E-3-0.68E-3)	0.60	-0.09E-3	(-0.89E-3-0.01E-3)	0.81	-0.41E-3	(-1.21E-3-0.39E-3)	0.31	-0.08E-3	(-0.90E-3-0.74E-3)	0.85
BSI <sub>theta</sub>	-0.54E-3	(-1.34E-3-0.25E-3)	0.18	-0.01E-3	(-0.67E-3-0.67E-3)	0.97	-0.49E-3	(-1.19E-3-0.20E-3)	0.16	-0.19E-3	(-0.90E-3-0.52E-3)	0.59
BSIdir	-0.36E-3	(-1.43E-3-0.71E-3)	0.51	-0.55E-3	(-1.46E-3-0.37E-3)	0.24	-0.85E-3	(-1.78E-3-0.07E-3)	0.07	-0.09E-3	(-1.03E-3-0.86E-3)	0.86
BSIdir <sub>delta</sub>	-0.11E-2	(-0.26E-2-0.03E-2)	0.11	-0.88E-3	(-2.09E-3-0.33E-3)	0.15	-0.13E-2	(-0.25E-2-0.04E-3)	0.04	-0.11E-2	(-0.24E-2-0.01E-2)	0.08
BSIdir <sub>theta</sub>	-0.53E-3	(-1.81E-3-0.75E-3)	0.42	-0.17E-3	(-1.27E-3-0.94E-3)	0.77	-0.73E-3	(-1.85E-3-0.40E-3)	0.20	-0.49E-3	(-1.6E-3-0.65E-3)	0.39

Regression coefficient ( $\beta$ ), 95% confidence interval (CI), p-value (P), Holm-Bonferroni corrected significance level:  $P < (0.05/\text{number of comparisons})$ , delta/alpha ratio (DAR), brain symmetry index (BSI), delta: mean power calculated over 1-4 Hz, theta: mean power calculated over 4-8 Hz, directional BSI (BSIdir), national institutes of health stroke scale (NIHSS), Fugl-Meyer motor assessment of the upper extremity (FM-UE).



Table 3.4 | Within- and between-subject associations between clinical and spectral characteristics

	Hybrid association models															
	NIHSS between-subject effects				NIHSS within-subject effects				FM-UE between-subject effects				FM-UE within-subject effects			
	$\beta$	95% CI	P	$\beta$	95% CI	P	$\beta$	95% CI	P	$\beta$	95% CI	P	$\beta$	95% CI	P	
DAR	0.23	(0.05-0.42)	0.02	0.10	(0.03-0.18)	0.007	-0.01	(-0.04-0.02)	0.46	-0.01	(-0.03-0.24E-2)	0.10	-0.01	(-0.03-0.24E-2)	0.10	
DAR <sub>ΔH</sub>	0.36	(0.13-0.60)	0.003	0.16	(0.06-0.26)	0.003	-0.02	(-0.05-0.02)	0.31	-0.02	(-0.04-0.15E-2)	0.07	-0.02	(-0.04-0.15E-2)	0.07	
BSI	0.74E-2	(0.32E-2-0.01)	<0.001	0.12E-2	(-0.11E-2-0.34E-2)	0.31	-0.04E-2	(-0.11E-2-0.03E-2)	0.22	-0.04E-2	(-0.07E2-0.03E-2)	0.33	-0.02E-2	(-0.07E2-0.03E-2)	0.33	
BSI <sub>delta</sub>	0.99E-2	(0.44E-2-0.02)	<0.001	0.12E-2	(-0.19E-2-0.44E-2)	0.44	-0.06E-2	(-0.15E-2-0.02E-2)	0.15	-0.06E-2	(-0.09E2-0.05E-2)	0.57	-0.02E-2	(-0.09E2-0.05E-2)	0.57	
BSI <sub>theta</sub>	0.79E-2	(0.27E-2-0.01)	0.004	0.05E-2	(-0.21E-2-0.31E-2)	0.70	-0.05E-2	(-0.13E-2-0.03E-2)	0.23	-0.05E-2	(-0.07E2-0.04E-2)	0.59	-0.02E-2	(-0.07E2-0.04E-2)	0.59	
BSIdir	0.02	(-0.41E-2-0.88E-2)	0.46	0.34E-2	(0.03E-2-0.72E-2)	0.07	-0.02E-2	(-0.11E-2-0.07E-2)	0.63	-0.02E-2	(-0.11E-2-0.03E-2)	0.006	-0.11E-2	(-0.19E-2-0.03E-2)	0.006	
BSIdir <sub>delta</sub>	0.02	(0.01-0.03)	<0.001	0.67E-2	(0.23E-2-0.01)	0.003	-0.18E-2	(-0.32E-2-0.04E-2)	0.01	-0.18E-2	(-0.22E-2-0.03E-2)	0.01	-0.12E-2	(-0.22E-2-0.03E-2)	0.01	
BSIdir <sub>theta</sub>	0.01	(0.11E-2-0.02)	0.03	0.45E-2	(0.07E-2-0.84E-2)	0.02	-0.09E-2	(-0.23E-2-0.05E-2)	0.21	-0.09E-2	(-0.17E-2-0.77E-4)	0.03	-0.09E-2	(-0.17E-2-0.77E-4)	0.03	

Regression coefficient ( $\beta$ ), 95% confidence interval (CI), p-value (P), Holm-Bonferroni corrected significance level:  $P < (0.05/\text{number of comparisons})$ , delta/alpha ratio (DAR), brain symmetry index (BSI), delta: mean power calculated over 1-4 Hz, theta: mean power calculated over 4-8 Hz, directional BSI (BSIdir), national institutes of health stroke scale (NIHSS), Fugl-Meyer motor assessment of the upper extremity (FM-UE).



BSI was positively associated with the NIHSS score ( $\beta=2.51 \cdot 10^{-3}$ ,  $P=0.01$ ; Table 3.3A). After correction for time, the association with NIHSS became stronger ( $\beta=4.52 \cdot 10^{-3}$ ,  $P<0.001$ ), which suggests an association between the dependent and independent covariates irrespective of the time-dependent changes of the covariates. The longitudinal relation mainly concerned a positive between-subject effect (Table 3.4). The interaction term between NIHSS and time did not reach significance, suggesting that the association between BSI and the NIHSS did not change over time. The  $BSI_{\text{delta}}$  and  $BSI_{\text{theta}}$  showed results similar to those for the BSI, yet remained borderline significant (Table 3.3A, 3.4).

$BSI_{\text{dir}_{\text{delta}}}$  showed a significant positive relation with NIHSS ( $\beta=0.84 \cdot 10^{-2}$ ,  $P<0.001$ ; Table 3.3A), where time was not a confounder. This relation consisted of significant positive within- and between-subject effects (Table 3.4). The relation between  $BSI_{\text{dir}_{\text{delta}}}$  and NIHSS was significant across measurement moments (Table 3.3B).

### Association between spectral characteristics and FM-UE

Tables 3.3A–C and 3.4 show the longitudinal associations between spectral characteristics and FM-UE. No significant longitudinal association was found between DAR or  $DAR_{\text{UH}}$  and FM-UE (Table 3.3A), neither when corrected for time nor at any specific moment in time (Table 3.3C). For  $DAR_{\text{AH}}$  a trend towards a negative association with FM-UE was found, which was no longer present after correction for time. This agrees with the outcome of the hybrid model, which revealed that this association was primarily caused by a within-subject effect (Table 3.4).

BSI,  $BSI_{\text{delta}}$  as well as  $BSI_{\text{theta}}$  did not show significant longitudinal associations with FM-UE (Table 3.3A), neither when corrected for time, nor at any moment in time (Table 3.3C).

$BSI_{\text{dir}}$  showed a trend towards a negative association with FM-UE ( $\beta=-0.72 \cdot 10^{-3}$ ,  $P=0.02$ ; Table 3.3A), but after correction for time this trend was no longer present. In line with this finding, the hybrid model showed only a significant negative within-subject effect (Table 3.4).  $BSI_{\text{dir}_{\text{delta}}}$  was negatively associated with FM-UE ( $\beta=-0.14 \cdot 10^{-2}$ ,  $P<0.001$ ; Table 3.3A), which was borderline significant after correction for time. This relation concerned significant negative within- and between-subject effects (Table 3.4). Further analyses revealed that the interaction term (FM-UE\*time) was significant, indicating that the relation varied over time.

## DISCUSSION

Current literature argues the importance of knowledge concerning the association between clinical improvements and changes in the brain after stroke<sup>28</sup>. Studies focused

on brain activity related to impairments after stroke mainly have a cross-sectional design. In the current study resting-state EEG and clinical data were measured repeatedly at recommended fixed moments in the first 6 months post-stroke<sup>28</sup>. The aim was to investigate longitudinal changes in the EEG-derived spectral characteristics DAR, BSI and BSIdir, as well as their changes over time in relation to improvements of NIHSS and FM-UE scores within and beyond the window of spontaneous neurobiological recovery.

We hypothesized that DAR, BSI and BSIdir would decrease mainly within the time window of spontaneous neurobiological recovery after stroke as reflected by changing neurological impairments such as FM-UE and NIHSS. However, our findings revealed that this time window did not fully match with the time window of changes in spectral characteristics, which were found to normalize within and beyond the first 3 months. In line with our second hypothesis, the time course of  $DAR_{AH}$  and  $BSIdir_{\text{delta}}$  within subjects was significantly positively associated with the severity of global neurological impairments as reflected by the NIHSS score. Moreover,  $BSIdir_{\text{delta}}$  showed a clear negative within-subject association with recovery of motor impairments of the upper extremity as reflected by FM-UE scores. This means that a decreasing asymmetry in the delta band within a patient was related with recovery of motor function of the upper extremity.

### Time course of spectral characteristics differs from spontaneous neurobiological recovery

Most of the spectral characteristics we investigated showed normalization over time to a certain extent, in line with what has been reported in the literature<sup>12,14,29</sup>. More specifically,  $DAR_{AH}$  and BSIdir approached values found in healthy subjects<sup>14</sup>, whereas lateralization as reflected by the BSI persisted. The seemingly inconsistent results for BSI and BSIdir might be the result of reciprocal asymmetries over the channel pairs, which accumulate in BSI while cancelling out in BSIdir. Nonetheless, our results show decreasing lateralization in the delta band for both asymmetry measures. Comparable with our results, a previous longitudinal MEG study reported delta activity to be increased in the affected hemisphere in the acute phase and to decrease over time during the early sub-acute phase post-stroke<sup>29</sup>.

The time course of the investigated spectral characteristics did not plateau at the same moment as spontaneous neurobiological recovery reflected by the NIHSS and FM-UE scores. This continuing normalization of EEG parameters suggests that not all changes measured with EEG reflect neurological improvements reflected by a global neurological deficits assessment as the NIHSS or motor function assessment as the FM-UE. Although speculative, the continuing normalization observed in EEG parameters may parallel more

refined neurological improvements, which are not detectable with NIHSS and FM-UE due to their ceiling effect. Obviously, molecular and cellular processes related to post-stroke recovery (i.e. upregulated growth factors, angiogenesis and synaptogenesis)<sup>7,30,31</sup> affect synaptic connections and network integrity, and lead to remapping<sup>5</sup>, which – in turn – may alter brain oscillations<sup>32</sup>. The underlying relationship between these processes remains to be investigated.

### Spectral characteristics as monitoring biomarkers of recovery

The positive within-subject effects found for DAR and  $DAR_{AH}$  reveal decreasing values within patients as NIHSS scores improved. This is in line with the findings of the aforementioned longitudinal MEG study, which revealed that patients with persistent low-frequency activity also had lower NIHSS scores than patients without such persistent low-frequency activity<sup>29</sup>.

The increased DAR values in both the affected and unaffected hemispheres, compared to healthy values, confirm the current literature reports suggesting that the unaffected hemisphere is also affected early after stroke<sup>33,34</sup>. Therefore, our asymmetry measure may have underestimated the neurological deficits early post-stroke. Since the unaffected hemisphere is less affected than the affected hemisphere,  $DAR_{AH}$  might be more appropriate to capture the relevant signals than DAR calculated over both hemispheres.

Our BSI results agree with those presented by Agius Anastasi and co-workers<sup>19</sup>. They reported a trend towards a decrease in BSI over time, and the absence of a significant correlation with FM-UE. We only showed a significant positive between-subject effect between NIHSS and BSI. This suggests that a lower NIHSS score in patient A compared to patient B, is related to a decreased BSI value in patient A compared to patient B.

The longitudinal associations between  $BSI_{dir_{\delta}}$ , stroke severity, and motor function as reported here emphasize the validity of this specification favoring the use of frequency bands and directionality. The within- and between-subject effects reveal that a lower degree of asymmetry in the delta band compared to another patient, or a decreasing degree of asymmetry in the delta band over time within a patient, were associated with decreased stroke severity and improved motor function of the upper extremity. This suggests that the development of  $BSI_{dir_{\delta}}$  and clinical scores over time within individuals are related.  $BSI_{dir_{\delta}}$  therefore shows potential as a monitoring biomarker of spontaneous neurobiological recovery.

In congruence with our data, which suggests increased activity towards the affected hemisphere in the delta frequency band (i.e. increased  $BSI_{dir_{\delta}}$ ), Fanciullacci and co-workers<sup>35</sup>

showed delta power to be increased in the affected compared to the unaffected hemisphere in stroke patients with subcortical lesions. Nonetheless, in the same sample they showed a negative correlation between pdBSI and NIHSS, which is different from our findings. This discrepancy may result from methodological issues such as small sample sizes in combination with the lack of correction for multiple testing. Hence, the influence of lesion location on these results has yet to be investigated. Other techniques and imaging methods (e.g. MRI or DTI) are necessary to better understand the impact of anatomical integrity on the time course of spectral characteristics early post-stroke.

### Limitations and future directions

Several limitations of the study should be taken into consideration. Additional analyses were performed in which the time courses of the spectral characteristics were compared between 3 patient groups classified based on their FM-UE recovery pattern (See Supplementary materials). Unfortunately, due to small subgroups this analysis was underpowered. Furthermore, since in the current study MRI data was unavailable for a large proportion of the patients, we were not able to correct for lesion size or location, while we acknowledge that this might influence the observed resting-state oscillations and motor recovery post-stroke<sup>35,36</sup>. In previous work, DAR was found to be only increased in patients with a cortico-subcortical lesion, while BSI was only increased in patients with a subcortical lesion when compared to healthy individuals<sup>35</sup>. Future studies are needed to further investigate the influence of lesion location on the time course of DAR and BSI. Additionally, we restricted the present study to spectral characteristics representing low-frequency activity. Whether alpha, beta and gamma frequencies may also be sensitive neurophysiological biomarkers of recovery early post-stroke needs to be investigated. Finally, due to the limited capacity of patients in the acute phase post-stroke, the baseline measurement took place at an average of 12 days post-stroke, which means that a substantial amount of recovery might already have occurred<sup>37</sup>. Hence, we may have missed some of the early changes in spectral characteristics over time.

In future research we suggest to investigate the contribution of low-frequency oscillations during upper limb movements. Previous work in rodents suggest that low-frequency oscillations are a possible target for neuromodulation to improve motor function recovery post-stroke<sup>38</sup>.

## Conclusion

In the current study, it was concluded that normalization of resting-state EEG asymmetry measures was not restricted to the time window of recovery of clinical neurological impairments measured with NIHSS and FM-UE. This might reflect an ongoing neural recovery beyond 3 months, which is not detectable by these impairment-focused outcome measures. In addition, global neurological recovery and recovery of motor function of the upper extremity are associated with normalization of their spectral characteristics in the low frequency bands in patients who suffered from ischemic stroke. Future research should investigate the influence of lesion location on this relationship as well as and the potential role of spectral characteristics as a prognostic biomarker of recovery.

## Acknowledgements

In addition to the authors of the present study, the consortium consists of Caroline Winters, Dirk Hoevenaars, Luuk Haring and Elza van Duijnhoven from Amsterdam UMC, location VUmc; Jun Yao and Julius Dewald from Northwestern University, Chicago; and Martijn Vlaar, Teodoro Solis-Escalante, Alfred Schouten, Yuan Yang, Mark van de Ruit, Konstantina Kalogianni, Joost van Kordelaar, Lena Filatova and Frans van der Helm from Delft University of Technology.

## Funding

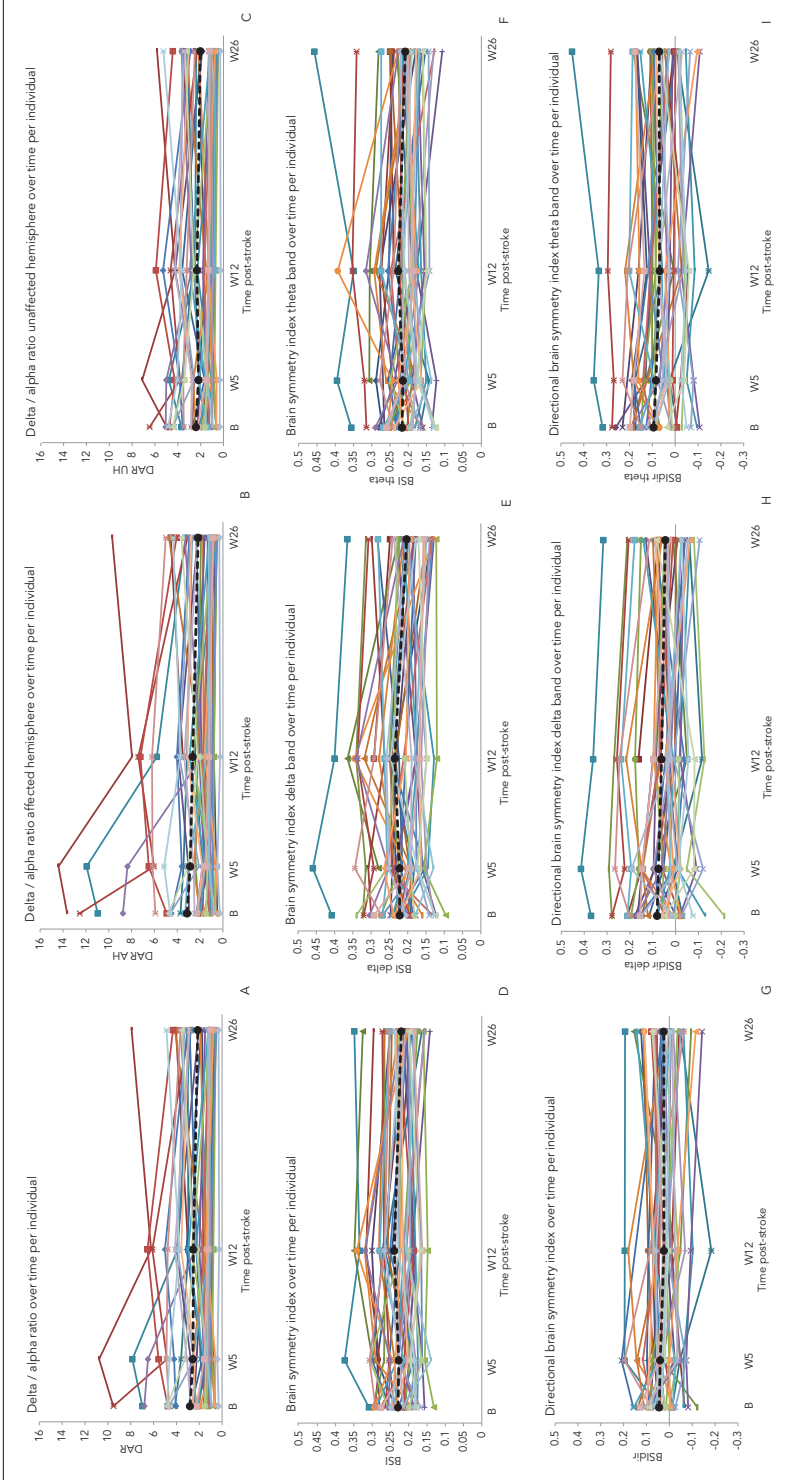
This study was financially supported by a European advanced grant (ERC-AG-ID1, FP7-IDEAS-ERC, 291339).

## REFERENCES

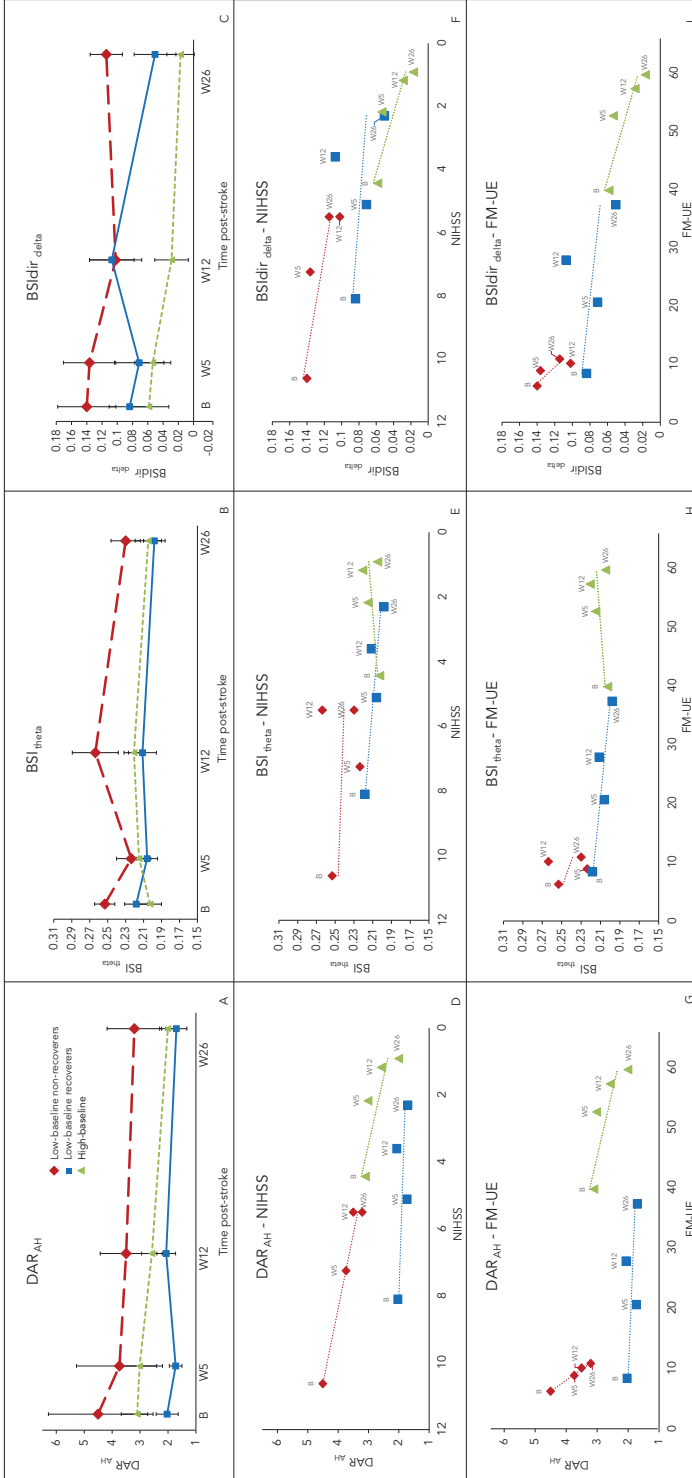
- 1 Langhorne P, Coupar F, Pollock A. Motor recovery after stroke: a systematic review. *Lancet Neurol.* 2009; **8**: 741–754.
- 2 Winters C, Van Wegen EEH, Daffertshofer A, Kwakkel G. Generalizability of the proportional recovery model for the upper extremity after an ischemic stroke. *Neurorehabil Neural Repair* 2015; **29**: 614–622.
- 3 Langhorne P, Bernhardt J, Kwakkel G. Stroke rehabilitation. *Lancet* 2011; **377**: 1693–702.
- 4 Buma F, Kwakkel G, Ramsey N. Understanding upper limb recovery after stroke. *Restor Neurol Neurosci* 2013; **31**: 707–722.
- 5 Murphy TH, Corbett D. Plasticity during stroke recovery: from synapse to behaviour. *Nat Rev Neurosci* 2009; **10**: 861–72.
- 6 Kwakkel G, Kollen B, Twisk J. Impact of time on improvement of outcome after stroke. *Stroke* 2006; **37**: 2348–2353.
- 7 Cramer SC. Repairing the human brain after stroke: I. Mechanisms of spontaneous recovery. *Ann. Neurol.* 2008; **63**: 272–287.
- 8 Buma FE, Lindeman E, Ramsey NF, Kwakkel G. Review: functional neuroimaging Studies of early upper limb recovery after stroke: a systematic review of the literature. *Neurorehabil Neural Repair* 2010; **24**: 589–608.
- 9 Ward N, Brown M, Thompson A, Frackowiak R. Neural correlates of motor recovery after stroke: a longitudinal fMRI study. *Brain* 2003; **126**: 2476–2496.
- 10 Boyd LA, Hayward KS, Ward NS, Stinear CM, Rosso C, Fisher RJ et al. Biomarkers of stroke recovery: consensus-based core recommendations from the stroke recovery and rehabilitation roundtable. *Int J Stroke* 2017; **12**: 480–493.
- 11 Ward NS. Restoring brain function after stroke - bridging the gap between animals and humans. *Nat Rev Neurol* 2017; **13**: 244–255.
- 12 Finnigan S, van Putten MJAM. EEG in ischaemic stroke: Quantitative EEG can uniquely inform (sub-)acute prognoses and clinical management. *Clin Neurophysiol* 2013; **124**: 10–19.
- 13 Van Putten MJAM, Tavy DLJ. Continuous quantitative EEG monitoring in hemispheric stroke patients using the brain symmetry index. *Stroke* 2004; **35**: 2489–2492.
- 14 Saes M, Meskers CGM, Daffertshofer A, de Munck JC, Kwakkel G, van Wegen EEH. How does upper extremity Fugl-Meyer motor score relate to resting-state EEG in chronic stroke? A power spectral density analysis. *Clin Neurophysiol* 2019; **130**: 856–862.
- 15 Bazanova O. Comments for Current Interpretation EEG Alpha Activity: A Review and Analysis. *J Behav Brain Sci* 2012; **02**: 239–248.
- 16 Britton J, Frey L, Hopp J, Korb P, Koubeissi M, Lievens W et al. *Electroencephalography (EEG): An Introductory Text and Atlas of Normal and Abnormal Findings in Adults, Children, and Infants.* 2016.
- 17 Sheerajpanday RVA, Nagels G, Weeren AJTM, van Putten MJAM, De Deyn PP. Reproducibility and clinical relevance of quantitative EEG parameters in cerebral ischemia: A basic approach. *Clin Neurophysiol* 2009; **120**: 845–855.
- 18 De Vos CC, Van Maarseveen SM, Brouwers PJAM, Van Putten MJAM. Continuous EEG monitoring during thrombolysis in acute hemispheric stroke patients using the brain symmetry index. *J Clin Neurophysiol* 2008; **25**: 77–82.
- 19 Agius Anastasi A, Falzon O, Camilleri K, Vella M, Muscat R. Brain symmetry index in healthy and stroke patients for assessment and prognosis. *Stroke Res Treat* 2017; **2017**: 8276136.
- 20 Tombaugh TN, McIntyre NJ. The Mini-Mental State Examination: A Comprehensive Review. *J Am Geriatr Soc* 1992; **40**: 922–935.
- 21 World Medical Association. World Medical Association declaration of Helsinki: Ethical principles for medical research involving human subjects. *JAMA - J Am Med Assoc* 2013; **310**: 2191–2194.
- 22 Zandvliet SB, van Wegen EEH, Campfens SF, van der Kooij H, Kwakkel G, Meskers CGM. Position-cortical coherence as a marker of afferent pathway integrity early post-stroke, a prospective cohort study. *Neurorehabil Neural Repair* 2020; **34**: 344–359.



- 23 Kwakkel G, Lannin NA, Borschmann K, English C, Ali M, Churilov L *et al.* Standardized measurement of sensorimotor recovery in stroke trials: Consensus-based core recommendations from the Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2017; **12**: 451–461.
- 24 Krakauer JW. Arm function after stroke: From physiology to recovery. *Semin. Neurol.* 2005; **25**: 384–395.
- 25 Gladstone DJ, Danells CJ, Black SE. The Fugl-Meyer Assessment of motor recovery after stroke: a critical review of its measurement properties. *Neurorehabil Neural Repair* 2002; **16**: 232–240.
- 26 Oostenveld R, Fries P, Maris E, Schoffelen J-M. FieldTrip: Open source software for advanced analysis of MEG, EEG, and invasive electrophysiological data. *Comput Intell Neurosci* 2011; **2011**: e156869.
- 27 Twisk JWR, de Vente W. Hybrid models were found to be very elegant to disentangle longitudinal within- and between-subject relationships. *J Clin Epidemiol* 2019; **107**: 66–70.
- 28 Bernhardt J, Hayward KS, Kwakkel G, Ward NS, Wolf SL, Borschmann K *et al.* Agreed definitions and a shared vision for new standards in stroke recovery research: The Stroke Recovery and Rehabilitation Roundtable taskforce. *Int J Stroke* 2017; **12**: 444–450.
- 29 Laaksonen K, Helle L, Parkkonen L, Kirveskari E, Mäkelä JP, Mustanoja S *et al.* Alterations in spontaneous brain oscillations during stroke recovery. *PLoS One* 2013; **8**: e61146.
- 30 Nudo RJ. Recovery after damage to motor cortical areas. *Curr. Opin. Neurobiol.* 1999; **9**: 740–747.
- 31 Cramer SC, Chopp M. Recovery recapitulates ontogeny. *Trends Neurosci.* 2000; **23**: 265–271.
- 32 Rabiller G, He JW, Nishijima Y, Wong A, Liu J. Perturbation of brain oscillations after ischemic stroke: A potential biomarker for post-stroke function and therapy. *Int J Mol Sci* 2015; **16**: 25605–25640.
- 33 Tecchio F, Pasqualetti P, Zappasodi F, Tombini M, Lupoi D, Vernieri F *et al.* Outcome prediction in acute monohemispheric stroke via magnetoencephalography. *J Neurol* 2007; **254**: 296–305.
- 34 Carter AR, Astafiev S V, Lang CE, Connor LT, Rengachary J, Strube MJ *et al.* Resting inter-hemispheric functional magnetic resonance imaging connectivity predicts performance after stroke. *Ann Neurol* 2010; **67**: 365–375.
- 35 Fanciullacci C, Bertolucci F, Lamola G, Panarese A, Artoni F, Micera S *et al.* Delta power is higher and more symmetrical in ischemic stroke patients with cortical involvement. *Front Hum Neurosci* 2017; **11**: 385.
- 36 Chen CL, Tang FT, Chen HC, Chung CY, Wong MK. Brain lesion size and location: Effects on motor recovery and functional outcome in stroke patients. *Arch Phys Med Rehabil* 2000; **81**: 447–452.
- 37 Winters C, Kwakkel G, Nijland R, Van Wegen E. When does return of voluntary finger extension occur post-stroke? A prospective cohort study. *PLoS One* 2016; **11**: e0160528.
- 38 Ramanathan DS, Guo L, Gulati T, Davidson G, Hishinuma AK, Won SJ *et al.* Low-frequency cortical activity is a neuromodulatory target that tracks recovery after stroke. *Nat Med* 2018; **24**: 1257–1267.



**Supplementary Figure S3.1 |** Individual and averaged (black dashed line) time course of the investigated spectral characteristics. Delta band: 1–4 Hz, theta band: 4–8 Hz. Abbreviations: baseline measurement within 3 weeks post-stroke (B), measurement week post-stroke (W), delta/alpha ratio (DAR), brain symmetry index (BSI), directional BSI (BSI<sub>dir</sub>), Fugl-Meyer motor assessment of the upper extremity (FM-UE), national institutes of health stroke scale (NIHSS).



Supplementary Figure S3.2 | Panel A–C: Time course of the spectral characteristics which showed trends in the groups analyses of the different recovery pattern groups: low-baseline non-recoverers (N=8, red dashed line, diamonds), low-baseline recoverers (N=10, blue solid line, squares) and high-baseline (N=23, green dotted line, triangles). Mean and standard errors of the mean are displayed. Panel D-i: Association between the EEG measures and NIHSS/FM-UE per group.

Abbreviations: Fugl-Meyer motor assessment of the upper extremity (FM-UE), national institutes of health stroke scale (NIHSS), baseline measurement within 3 weeks post-stroke (B), measurement week post-stroke (W), delta/alpha ratio of the affected hemisphere (DAR<sub>AH</sub>), brain symmetry index of the theta frequency band (BSI<sub>theta</sub>), directional BSI of the delta frequency band (BSI<sub>dir\_delta</sub>).







# Chapter 4

Poor motor function is associated with reduced sensory processing after stroke

S. Floor Campfens  
Sarah B. Zandvliet  
Carel G.M. Meskers  
Alfred C. Schouten  
Michel J.A.M. van Putten  
Herman van der Kooij

*Experimental Brain Research* 2015;  
233(4): 1339–1349

## ABSTRACT

The possibility to regain motor function after stroke depends on the intactness of motor and sensory pathways. In this study, we evaluated afferent sensory pathway information transfer and processing after stroke with the coherence between cortical activity and a position perturbation (position-cortical coherence, PCC).

Eleven subacute stroke survivors participated in this study. Subjects performed a motor task with the affected and non-affected arm while continuous wrist position perturbations were applied. Cortical activity was measured using EEG. PCC was calculated between position perturbation and EEG at the contralateral and ipsilateral sensorimotor area.

The presence of PCC was quantified as the number of frequencies where PCC is larger than zero across the sensorimotor area. All subjects showed significant contralateral PCC in affected and non-affected wrist tasks. Subjects with poor motor function had a reduced presence of contralateral PCC compared with subjects with good motor function in the affected wrist tasks. Amplitude of significant PCC did not differ between subjects with good and poor motor function. Our results show that poor motor function is associated with reduced sensory pathway information transfer and processing in subacute stroke subjects. Position-cortical coherence may provide additional insight into mechanisms of recovery of motor function after stroke.



## INTRODUCTION

Stroke is a leading cause of adult onset disability in the Western world. Rehabilitation after stroke has a strong emphasis on reducing motor impairment to improve the quality of life <sup>1</sup>. Within rehabilitation practice, sensory impairment does not receive as much attention as motor impairment does, although it is known that sensory impairment is common after stroke <sup>2</sup> and related to motor impairment <sup>3</sup>.

The relation between sensory and motor impairment is unsurprising because motor control requires bidirectional interaction between cortex and periphery <sup>4,5</sup>. Sensory feedback via the afferent pathways is necessary to generate proper motor commands which reach the muscles via the efferent pathways. Invasive recordings in monkeys showed that both sensory and motor cortical neural populations synchronise their oscillatory activity to peripheral signals <sup>6</sup>. This synchronisation is thought to play an important role in the transmission of information, i.e. connectivity, within closed loop motor control <sup>4,7,8</sup>. Coherence between cortical activity and muscle activity, corticomuscular coherence (CMC) <sup>9-11</sup>, is used as a measure of cortical sensorimotor integration during a motor task and depends on the information transfer across both efferent and afferent pathways <sup>12-15</sup>.

As a measure of both efferent and afferent pathway connectivity, changes in CMC cannot be related to changes in sensory or motor pathways. In addition, measurement of CMC requires a measurable EMG signal and thus is only possible in subjects that are able to voluntarily generate muscle force. When studying sensory and motor function after stroke with CMC, no information can be obtained from individuals without voluntary muscle control. Finally, a large downside for the potential clinical application of CMC is that it cannot be detected in all cases: even healthy subjects, with normal voluntary motor control, do not all present CMC <sup>16-18</sup>. The inter-individual difference in the presence of CMC reflects physiological inter-individual differences in the strength of the oscillatory corticomuscular coupling and is not the result of technical aspects such as the (mis-) placement of EEG electrodes <sup>17</sup>.

We previously showed that adding a small continuous position perturbation during an isotonic force task elicits CMC and coherence between the position perturbation and the EEG: position-cortical coherence (PCC) <sup>16</sup>. Position-cortical coherence represents the unidirectional information transfer across the afferent pathways because the perturbation acts as an external excitation signal for the proprioceptive system (primarily the Golgi tendon organs and muscle spindles). Coherence between position perturbation and cortical activity can only occur when sensory feedback related to the perturbation reaches the cortex via the afferent pathways: information transfer. Possibly, also neural populations

that are involved in the processing and sensorimotor integration of this information synchronise their activity to the perturbation as well and contribute to PCC. Position-cortical coherence thus represents sensory information transfer and processing. The origin of PCC is comparable to the origin of steady state evoked potentials of the visual or auditory system <sup>19</sup>. Because PCC is present in all subjects, it is a more reliable measure to study the cortical involvement in motor control than CMC <sup>16</sup>. In addition, as PCC represents unidirectional connectivity it has a simpler interpretation than CMC which represents bidirectional connectivity and does not allow estimation of pathway specific properties <sup>20</sup>.

The aim of this study is to show the potential value of PCC as a measure of sensory pathway information transfer and processing after stroke. Based on the association between sensory and motor impairment after stroke <sup>3</sup>, we hypothesise that stroke survivors with poor motor function have a lower PCC. We made a distinction between the presence of PCC (i.e. the number of frequencies and electrodes with significant PCC) and the mean amplitude of significant PCC in order to assess which is most informative of sensory pathway information transfer and processing.

In addition, we introduce a lateralisation index of PCC to evaluate the distribution of PCC between the lesioned and non-lesioned hemisphere. While in normal subjects PCC was localised at the contra-SM <sup>16</sup>, it has been shown with fMRI and EEG that stroke survivors recruit additional, ipsilateral areas during movement <sup>21,22</sup>. We therefore hypothesise that stroke survivors present PCC in both hemispheres.

## METHODS

Eleven first-ever hemispheric stroke survivors participated in the study (one woman). Details of the subjects are presented in Table 4.1; all subjects had ischaemic stroke. Subjects were either outpatients or recruited in the acute phase from the hospital wards of the Leiden university medical center. Subjects were in the subacute phase after stroke (within 6 months post-stroke). The group of subjects had a dichotomous distribution on the Fugl-Meyer motor assessment of the upper extremity (FM-UE) <sup>23</sup>. Six subjects had a FM-UE of 55 points or higher (the maximum possible score is 66 points), these subjects were considered to have a good motor function (group: good function). A score of 55 or higher indicates that these subjects were able to move outside of synergistic movement patterns. Some of these patients still experienced loss of strength and coordination, indicated by a non-maximal score; however, they were able to perform all movements and withstand a minimal resistance during the test. The other 5 subjects had considerably lower FM-UE scores, these subjects all scored <20 points and were considered to have

Table 4.1 | Overview of subjects

Subject code	Age	Group	Lesion	Days since lesion	FM-UE	MAS wrist	Sensory function (EmNSA-UE)	Task affected arm <sup>a</sup>
C.01	77	Good function	MCA left	106	65	0	Normal	Active
C.02	58	Good function	BG right	36	63	0	Normal	Active
C.06	77	Good function	MCA right	18	56	0	Normal	Active
C.08	35	Good function	Thal. right	22	63	0	Normal	Active
C.09	54	Good function	MCA left	60	65	0	Normal	Active
C.11	72	Good function	MCA right	13	65	0	Normal	Active
C.04	62	Poor function	MCA right	27	17	1	Normal	Relax
C.05	59	Poor function	MCA right	21	15	1+(flex)	Normal	Relax
C.07	58	Poor function	MCA right	11	19	0	Recued tactile	Relax
C.10	46	Poor function	MCA right	34	6	1+(flex)	Normal	Relax
C.13	67	Poor function	MCA right	121	6	4(flex)	Reduced tactile and proprioception	Relax

Abbreviations: middle cerebral artery (MCA), basal ganglia (BG), thalamus (Thal), Fugl-Meyer motor assessment of the upper extremity (FM-UE), modified ashworth scale (MAS), Erasmus modification of the Nottingham sensory assessment of the upper extremity (EmNSA-UE).

<sup>a</sup> All subjects performed the active task with the unaffected arm.

poor motor function (group: poor function). Sensory function of subjects was evaluated using the Erasmus modification of the Nottingham sensory assessment (EmNSA)<sup>24</sup>. This scale evaluates different test items across multiple locations on the upper extremity. Test items are as follows: light touch, pressure, pin prick, sharp–blunt discrimination, and proprioception. When a subject scored less than the maximal score for a test item on more than one location on the upper extremity, the test item is marked as reduced.

All measurements were taken in accordance with the *Declaration of Helsinki* and were approved by the medical ethics review committee of the Leiden university medical center (Leiden, the Netherlands). All participants gave signed informed consent before the measurements.

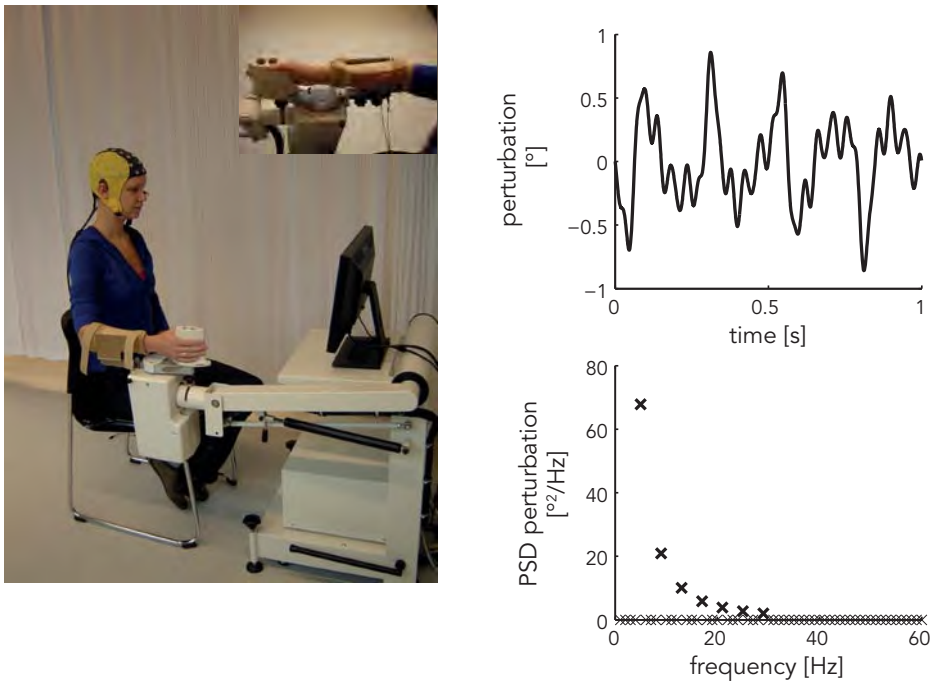
### Experimental set-up

Subjects were seated next to a wrist manipulator (Moog Inc., Nieuw-Vennep, the Netherlands), see Figure 4.1. The wrist manipulator (WM) is an actuated rotating device with a single degree of freedom that can impose flexion and extension movements on the wrist. The lower arm of the subject was strapped in an arm rest while the subject held the handle of the WM. The axis of rotation of the WM was aligned with the axis of rotation of the wrist. The neutral angle was determined for each subject as an angle between flexion and extension which was comfortable for the subject. The lever of the WM is equipped with a force transducer to measure the torques exerted by the subject.

EEG was measured using 64 scalp electrodes, placed according to the 5% electrode system<sup>25</sup> using a standard EEG cap with Ag/AgCl electrodes (actively shielded headcap by TMSi, Oldenzaal, the Netherlands). Electrode impedances were below 20 k $\Omega$ , and signal quality was monitored online. EMG was measured from the flexor carpi radialis using bipolar Ag/AgCl electrode pairs placed on the muscle belly. All physiological signals were sampled at 2048 Hz (Refa system by TMSi, Oldenzaal, the Netherlands). The angle of the WM and the torque exerted on the lever were synchronously measured on a separate system at 2048 Hz (Porti system by TMSi, Oldenzaal, the Netherlands) or via electrically isolated channels on the same amplifier as the physiological signals.

### Protocol

Subjects performed a motor task with the affected arm and with the non-affected arm. In the active motor task, subjects were instructed to exert a constant wrist flexion torque on the handle of the WM. Subjects were instructed to keep the exerted torque within a range of  $1.8 \pm 0.27$  Nm. To aid the subjects in keeping a steady torque, subjects received



**Figure 4.1** | Overview of the experimental set-up (left) and a 1-s segment of the position perturbation (right).

The subject holds the handle of the wrist manipulator (WM), and the lower arm is strapped in an arm rest using Velcro straps. To support the subject in maintaining a steady isotonic wrist flexion torque, visual feedback of the target torque and the exerted (2-Hz low-pass filtered with third-order Butterworth filter) was provided on the display in front of the subject. The position perturbation is a sum of sines with a decreasing value of the power with frequency. Power spectral density (PSD).

visual feedback of the exerted and the target torque via a display. For the visualisation, the exerted torque was low-pass filtered online with a cut-off frequency below the bandwidth of the perturbation (third-order low-pass Butterworth, 2 Hz) to remove frequencies contained in the position perturbation.

All subjects performed the active motor task (active task) with the non-affected arm and attempted the active task with the affected arm. When a subject was unable to maintain a stable wrist flexion torque for at least 5 s with the affected wrist or when the subject was unable to return to the target torque once the exerted torque decreased, the subject performed the relax task. In the relax task, the subject held the handle of the WM without exerting a torque. Table 4.1 lists which task was performed with the affected arm for each subject. Subjects performed eight trials of 40 s. In case the online monitoring of the

EEG revealed many eye blinks or activation of facial muscles, an extra trial was recorded. The affected arm trials were performed first. During the trials, the inactive hand lay in a comfortable relaxed position, generally on the lap of the subject. EMG was visually monitored online to control for mirror movements.

Six subjects (C.05, C.07, C.08, C.10, C.11, and C.13) were willing to perform both the active and relax task with the affected and/or non-affected arm to allow comparison between the active and relax task. Two subjects performed these extra trials in a separate measurement session. The subjects with poor motor function that performed the extra active motor tasks with the affected arm (C.05, C.07, and C.13) performed the extra active task with a lower target torque. The target torque was set such that muscle activation was seen in the EMG signal. Even with a low target torque, these subjects were unable to maintain a stable contraction; during the trials, subjects were motivated to keep attempting to exert torque and return to the target torque when the exerted torque decreased.

The position perturbation signal (Figure 4.1, right side) consisted of a sum of sine waves (5, 9, 13, 17, 21, 25, and 29 Hz). The perturbation signal had a period of 1s and a peak-to-peak amplitude of  $1.7^\circ$  (0.03 rad). The power of the sine waves decreased with frequency, giving the perturbation a flat velocity spectrum. The small amplitude of the perturbation allows the application of the perturbation also to subjects with increased wrist stiffness.

### Data analysis

Recorded signals were processed offline using MATLAB 2010b (the MathWorks, Inc., Natick, MA, USA). First, raw EEG signals were high-pass filtered (1 Hz, second-order Butterworth filter applied with zero phase shift) to remove baseline drift. Channels containing artefacts due to bad electrode contact were removed from the common average reference. EEG signals were low-pass filtered (70 Hz, second-order Butterworth applied with zero phase shift) and resampled to 1024 Hz.

All signals were segmented in 1 s segments (1024 samples), the period of the perturbation, with 75% overlap between segments. Segments were visually inspected, and segments that contained eye blinks or muscle activity were removed. The 50 Hz component was removed from each segment using the discrete Fourier transform<sup>26</sup>. EEG data were then referenced to a nearest neighbour Laplacian derivation.

Subsequent coherence analysis was performed on EEG channels overlying the left and right sensorimotor areas (SM). The left SM consists of FC1, FC3, FC5, C1, C3, C5, CP1, CP3, and CP5, and the right SM consists of the equivalent electrodes on the right hemisphere.

## Estimation of PCC

All segments were transformed to the frequency domain using the fast Fourier transform. The power spectral density (PSD,  $\phi_{xx}(f)$ ) and cross-spectral density (CSD,  $\phi_{xy}(f)$ ) were estimated using:

$$\Phi_{xx}(f) = \frac{1}{N} \sum_{i=1}^N X_i^*(f) \cdot X_i(f) \quad (1)$$

and

$$\Phi_{xy}(f) = \frac{1}{N} \sum_{i=1}^N X_i^*(f) \cdot Y_i(f) \quad (2)$$

respectively, where  $X_i(f)$  and  $Y_i(f)$  are the Fourier coefficients at frequency  $f$ , estimated from the  $i_{th}$  data segment. The asterisk indicates the complex conjugate, and  $N$  is the total number of segments.

EEG channels were excluded from coherence analysis when the mean power in the frequency band between 25 and 49 Hz was larger than the mean power between 5 and 15 Hz. Channels with this power distribution were presumed to reflect mostly EMG activity. This method for marking channels with EMG activity is based on the method applied by Severens *et al.*<sup>27</sup> to select EEG and EMG components in a blind source separation method. The presence of EMG activity obscures the detection of PCC as it severely decreases signal to noise ratio. The (magnitude-squared) coherence ( $C_{xy}(f)$ ) between signals was calculated according to:

$$C_{xy}(f) = \frac{|\Phi_{xy}(f)|^2}{\Phi_{xx}(f)\Phi_{yy}(f)} \quad (3)$$

Position-cortical coherence (PCC) was calculated between the position perturbation signal and each EEG channel and was only evaluated at the frequencies contained in the perturbation signal. The significance of coherence values was determined using the approximation of the confidence limit (CL) by Bortel and Sovka<sup>28</sup>. The confidence level was set to 0.99 ( $\alpha=0.01$ ).

The presence of PCC across the sensorimotor area contralateral to the wrist perturbation (contra-SM) was evaluated by summing the number of frequencies where the PCC exceeds the 99% CI per electrode and summing across the contra-SM. This number was expressed as a percentage of the total number of frequency bins on the contra-SM (i.e. number of stimulus frequencies times the number of electrodes in the contra-SM). Amplitude of significant PCC was evaluated by the mean significant PCC over the contra-SM.



Lateralisation of PCC was quantified by the lateralisation index (L):

$$L = \log_{10}(PCC_{\text{contra-SM}}) - \log_{10}(PCC_{\text{ipsi-SM}}) \quad (4)$$

where  $PCC_{\text{contra-SM}}$  and  $PCC_{\text{ipsi-SM}}$  are the mean PCC amplitudes over all frequencies and all electrodes in the contra-SM and ipsilateral SM (ipsi-SM), respectively. Note that in the lateralisation index, no distinction is made between the presence and amplitude of significant PCC. When  $L > 0$ , the PCC is more lateralised towards the contra-SM.  $L < 0$  indicates that PCC is more lateralised towards the ipsi-SM.

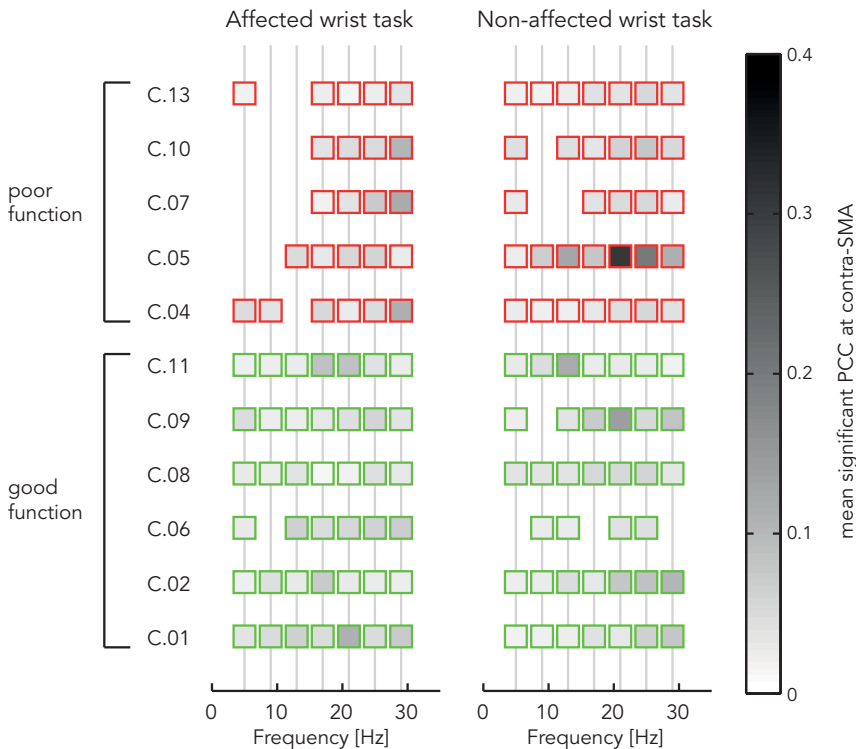
The nonparametric Wilcoxon rank-sum test was used to compare the presence and amplitude of significant PCC at the contra-SM and the lateralisation index between the good function and poor function subjects. Paired t tests were used to compare contra-SM PCC and lateralisation index in response to affected wrist perturbation and non-affected wrist perturbation within one subject. Amplitude of significant PCC was log10-transformed prior to statistical analysis.

## RESULTS

All good function subjects had normal sensory function on all test items according to the EmNSA scale (see Table 4.1). In the poor function group, C.07 had a reduced sensory function affected arm at the time of the first measurement session (11 days post-stroke). C.07 had a reduced tactile sensation in the hand and fingers and a reduced ability to discriminate between sharp and blunt tactile stimuli in the whole arm including hand and fingers. In addition, C.13 had a reduced sensory function in the affected arm: a reduced ability to discriminate between sharp and blunt stimuli across the whole arm including hand and fingers and a reduced proprioceptive sense in the fingers, wrist, and elbow. In all 11 subjects, EEG on one or more electrodes in the left and right SM was excluded due to poor signal quality. In 9 subjects, this concerned one or more of the most temporal electrodes (FC5, FC6, C5, C6, CP5, and CP6). In 2 subjects, also electrodes other than the most temporal ones were excluded. For C.06, electrodes FC3, C3, and CP3 were excluded in addition to the most temporal electrodes (FC5, FC6, C5, C6, CP5, and CP6). For C.07, electrodes C3 and FC3 were excluded in addition to the most temporal electrodes (FC5, FC6, C5, and C6).

### Presence and amplitude of significant PCC in the contralateral sensorimotor area

All subjects presented significant PCC at the contra-SM on at least three stimulus frequencies in the affected wrist task and in the non-affected wrist task (see Figure 4.2). Five of the 6 subjects in the good function group had significant contra-SM PCC during the affected wrist task on all stimulus frequencies, while none of the subjects in the poor function group had significant contra-SM PCC on all stimulus frequencies during the affected wrist task. The poor function subjects all presented significant PCC on the highest stimulus frequencies (17, 21, 25, and 19 Hz) in the affected wrist task.



**Figure 4.2** | Mean significant PCC at the contra-SM per stimulus frequency.

Left: affected wrist task, right: non-affected wrist task. Grey vertical lines indicate the stimulus frequencies, a square indicates that the PCC was significantly larger than zero at that stimulus frequency on at least one electrode in the contra-SM.

The presence of significant PCC at the contra-SM varied between affected and non-affected wrist tasks and between subjects (Figure 4.3). Poor function subjects tended to have a lower presence of contra-SM PCC in the affected wrist task compared with the non-affected wrist task but the difference did not reach statistical significance. The difference in the

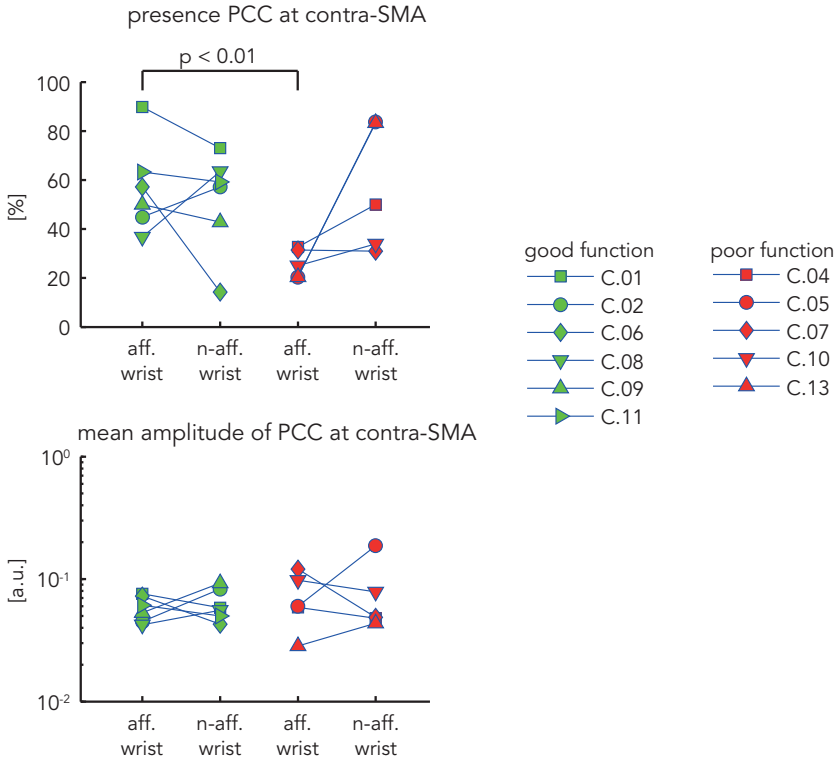


Figure 4.3 | Presence and amplitude of significant PCC at the contra-SM in the affected wrist task (aff. wrist) and non-affected task (n-aff. wrist).

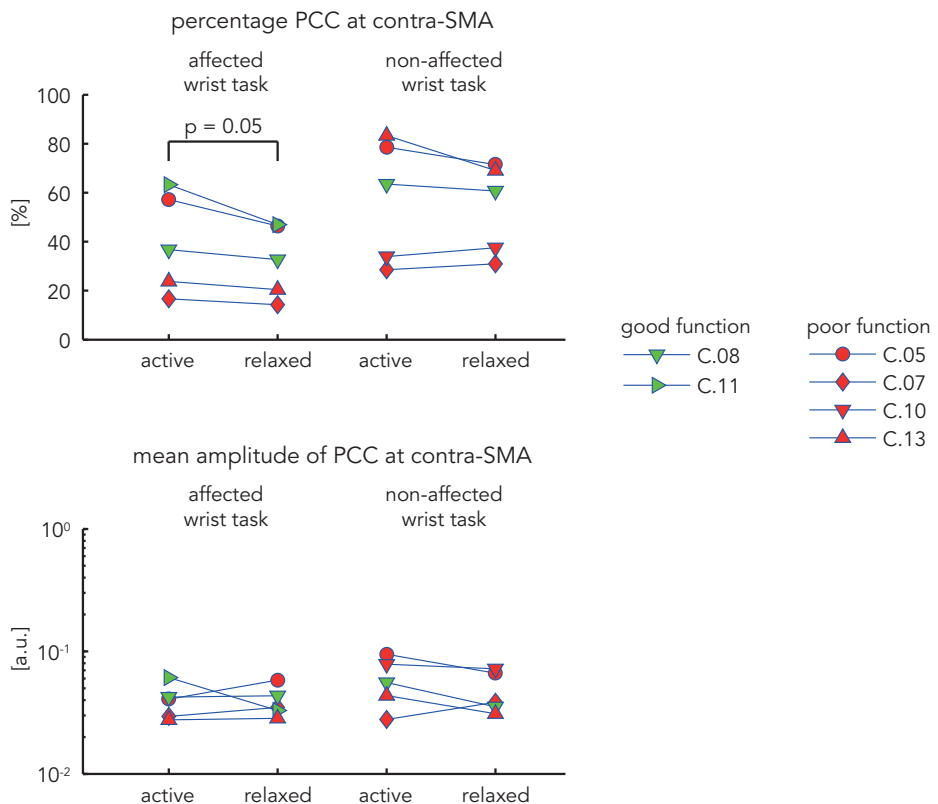
Upper panel: Presence of PCC expressed as a percentage of the total number of frequency bins on the contra-SM. Lower panel: Mean significant PCC over the contra-SM and all stimulus frequencies in arbitrary units (a.u.).

presence of contra-SM PCC in the affected wrist tasks between poor and good function subjects was significant (Wilcoxon rank-sum test:  $P < 0.01$ ). The average difference between the subject groups was 31%.

The mean significant amplitude of PCC on the contra-SM varied between subjects and between affected and non-affected wrist tasks (Figure 4.3). There was no significant difference in mean PCC amplitude at contra-SM between good and poor function subjects, not in the affected wrist task and not in the non-affected wrist task. Neither was there a significant difference between contra-SM PCC amplitudes in affected and non-affected wrist tasks within subjects.

## Comparison of PCC in active and relaxed tasks

The difference in tasks performed with the affected wrist by the subjects with poor function and those with good motor function (relax and active motor tasks, respectively) could be a confounding factor. Therefore, 5 subjects performed an extra active or relax task with the affected and/ or non-affected wrist to enable comparison of the presence and mean significant amplitude of contra-SM PCC between the active and relax tasks. Results are summarised in Figure 4.4. The presence of contra-SM PCC in the affected wrist tasks tended to be lower in the relax task compared with the active task. This difference was on the limit of significance (paired t test:  $P=0.05$ ). The average difference in the presence of contra-SM PCC was 7.4%. In the non-affected wrist tasks, the difference was not significant. There were no significant differences in mean significant contra-SM PCC amplitude between active and relaxed tasks.



**Figure 4.4** | Comparison of the presence and magnitude of significant PCC between an active and a relaxed task.

Upper panel: Presence of PCC expressed as a percentage of the total number of frequency bins across the contra-SM. Lower panel: Mean significant PCC over the contra-SM in arbitrary units (a.u.). C.10 performed both tasks only with the non-affected wrist, and C.11 performed both tasks only with the affected wrist.

## Lateralisation of PCC

In ten subjects, PCC was significantly larger than zero on at least one electrode on the ipsi-SM during both the affected and the non-affected wrist task. Only C.10 did not show significant PCC on the ipsi-SM during the non-affected wrist task, and thus, the lateralisation index could not be determined in this case. In the non-affected wrist task, the lateralisation index was larger than zero in all subacute subjects, showing that PCC was lateralised more towards the contra-SM (see Figure 4.5).

In the poor function group, the lateralisation index was significantly lower during the affected wrist task compared with the non-affected wrist task (paired t test:  $P=0.03$ ). This indicates that in the poor function group, PCC is distributed more evenly between the lesioned and non-lesioned hemisphere during the affected wrist task, while during the non-affected wrist task PCC is more lateralised to the (non-lesioned) contra-SM.

Three subjects (C.06, C.05, and C.13) had a negative lateralisation index during the affected wrist task. This indicates that the PCC is more lateralised towards the ipsi-SM.

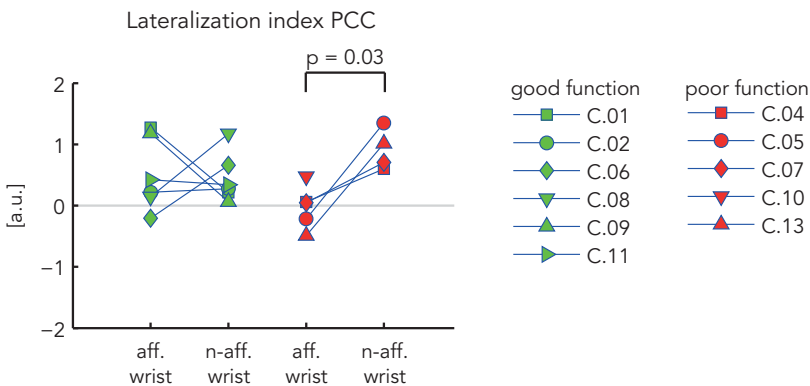


Figure 4.5 | Lateralisation index for affected and non-affected wrist tasks.

$L > 0$  indicates that PCC is more lateralised towards the contra-SM compared with the ipsi-SM.  $L < 0$  indicates that PCC is more lateralised towards the ipsi-SM.

affected (aff), non-affected wrist (n-aff wrist), in arbitrary units (a.u.).

## DISCUSSION

In this study, we evaluated position-cortical coherence (PCC) in eleven subacute stroke subjects. Position-cortical coherence is the coherence between a joint position perturbation and the EEG during an isotonic motor task. Because the position perturbation acts as an independent external excitation of the proprioceptive system, PCC represents unidirectional connectivity, i.e. information transfer, across the afferent pathways<sup>16</sup>. All stroke subjects

presented PCC at the contralateral sensorimotor area (contra-SM) during an affected and non-affected wrist angle perturbation. The presence of contra-SM PCC during an affected wrist perturbation was lower in poor motor function stroke subjects than in good motor function stroke subjects. The presence of contra-SM PCC differed between subjects with good and poor motor function; the amplitude of significant contra-SM PCC did not differ between these groups. This implies that when contra-SM PCC is significantly larger than zero, the amplitude does not contain extra information. The reduced presence of PCC in the ipsilesional hemisphere of poor function subjects shows that these subjects have not only poor functioning efferent motor pathways, but a reduced integrity of their afferent sensory pathways as well. This agrees with the notion that motor control takes place in a closed loop where sensory feedback is crucial for generating proper motor commands.

From the reduced presence of PCC, it cannot directly be determined whether subjects with poor motor function showed contra-SM PCC on less frequencies or on less electrodes. Damage to the sensorimotor cortex due to a large cortical stroke can result in a reduction in the size of the sensorimotor cortex and thus PCC on less electrodes. However, none of the poor function subjects presented significant PCC on the lowest stimulus frequencies in the affected wrist task (Figure 4.2). This indicates that a reduced number of frequencies where PCC is present is a large contributing factor to the reduced presence of PCC in the poor function subjects. Future studies combining PCC measurements with detailed imaging of the cortex should correlate the presence of PCC to specific (cortical) damage.

In most data sets, electrodes had to be excluded because the EEG signals were contaminated by muscle activity. As a result, we may have missed significant PCC. For future studies, it is advised to consider the use of artefact removal algorithms to remove muscle activity artefacts from the EEG. A potentially suitable algorithm based on blind source separation was presented by De Vos *et al.*<sup>29</sup> for the removal of tonic muscle activity from EEG recorded during spoken language.

In the group of subacute stroke subjects included in this study, the subjects with good and poor motor function had very distinct ranges of FM-UE scores. Due to this sharp distinction, we cannot correlate the PCC measures with the FM-UE score. In a larger population of stroke subjects, that exhibit a more continuous distribution of FM-UE scores, it would be possible to perform correlation analysis on the presence of contra-SM PCC and motor function score to find out whether there is a continuous relation between motor function and afferent pathway integrity.

Although the presence of contra-SM PCC in the affected wrist task was lower in the poor function group, most subjects in the poor function group had a normal sensory

function according to the EmNSA<sup>24</sup>. Subject C.13 had a reduced proprioceptive sense of the fingers, wrist, and elbow but a presence of PCC similar to the other subject in the group. This implies that sensory feedback related to the perturbation from the muscle spindles and Golgi tendon organs does reach the cortex. Possibly the sensory feedback is processed in a different, less effective, manner at the cortical level, resulting in the reduced sensory function according to the clinical assessment of this subject. However, as the other subjects with poor motor function had normal sensory function according to the clinical assessment, the lower presence of PCC in the poor function group could suggest that sensory deficits in this group were not found with the clinical assessment. Clinical scaling of sensory function has been shown to be unreliable<sup>30</sup>. Although progress has been made to obtain more reliable clinical scoring of sensory function<sup>24</sup>, especially impairment of proprioception is often overlooked<sup>31</sup>. Dukelow *et al.* introduced a method for the assessment of proprioceptive function using robotics. Such robotic assessment of sensory function could very well be combined with an evaluation of PCC to enable a more quantitative evaluation of the relation between PCC and proprioceptive function.

Subjects with poor motor function were not able to generate a steady wrist flexion torque, and these subjects performed the relax task. Also during the relax task, subjects showed significant contra-SM PCC. In the subjects that performed additional active and relax tasks for comparison, the presence of contra-SM PCC did not differ between active and relax tasks performed with the non-affected wrist. This implies that the cortical response to the position perturbation is similar during active and relax tasks. However, when performed with the affected wrist the presence of PCC was 7% smaller during a relax task. This difference between active and relax tasks is not sufficient to explain the difference in the presence of PCC found between the subjects with good and poor motor function (30%). Although the difference between active and relax tasks is small or absent, we advise to let subjects relax in future studies to fully avoid biased results due to differences in ability to perform motor tasks.

The frequency-dependent likelihood of finding significant contra-SM PCC during the affected wrist task in the poor function group indicates that there is a frequency dependency of the signal to noise ratio in the ipsilesional EEG. Either the EEG contains more contributions of sources other than the position perturbation at the lowest stimulus frequencies (increased noise at lowest stimulus frequencies) or the contribution of the position perturbation to the EEG is higher at the highest stimulus frequencies (increased signal at the highest stimulus frequencies). The perturbation signal was designed such that it had a flat velocity spectrum. As a result, the position- and velocity-sensitive muscle spindles were excited with decreasing and equal power at each frequency, respectively.



Higher sensitivity of muscle spindles to higher velocities or excitation of Golgi tendon organs could still result in a higher output of the sensory pathways at the higher stimulus frequencies, thus increasing the likelihood of finding PCC at these frequencies. An alternative explanation for an increased signal at the highest stimulus frequencies could be that these stimulus frequencies are transmitted more efficiently along the afferent pathways. The higher stimulus frequencies lie in the beta band (15–30 Hz). During an isotonic and isometric motor task, coherence between EEG and EMG is typically found in the beta band, indicating that oscillations in this frequency band are already being transmitted along the afferent pathways.

Using the lateralisation index of PCC, we found that 3 of the 11 subjects (2 poor function and 1 good function) had PCC predominantly localised in the ipsi-SM during the affected wrist task. It is known from several studies that after stroke, there can be a disbalance between the lesioned and the non-lesioned hemisphere. This results in abnormal activation of the ipsilateral sensorimotor cortex during motor tasks with the affected hand and is especially seen in stroke survivors with poor recovery of motor function<sup>21,22</sup>. During motor tasks with the affected side, the increased activity of the ipsilateral sensorimotor cortex is seen as a sign of increased motor output of the ipsilateral sensorimotor cortex. Our results in lateralisation of PCC indicate that sensory input from the affected wrist elicits response in the ipsilateral sensorimotor cortex and that this response can even be stronger than in the contralateral sensorimotor cortex. Increased activation of the ipsilateral sensorimotor cortex can thus also indicate increased sensory input to this area during an affected arm motor task. Further research is required to establish whether this relates to recovery of motor function.

About two third of the stroke survivors with initial hemiplegia do not regain dexterity and remain impaired despite rehabilitation therapy<sup>32,33</sup>. The ability to extend the fingers and abduct the shoulder at 72 h post-stroke gives a strong indication of an individual's ability to recover motor function<sup>34</sup>. Nevertheless, 25% of the stroke survivors with an initial poor prognosis does regain dexterity. It is important to identify individuals in this so-called crossover group since they will most likely benefit most from early applied intensive rehabilitation training. Potentially, objective assessment of connectivity between cortex and periphery could aid in identifying individuals in the crossover group. Position-cortical coherence may be more suitable for this purpose because it has important advantages over CMC, which is the traditional measure of corticomuscular connectivity<sup>16</sup>. While CMC cannot be found in all subjects, all subjects do present PCC. Furthermore, PCC can be measured even in subjects that lack the ability to voluntarily generate muscle contraction, while muscle activation measurable by EMG is required for the estimation

of CMC. In a larger longitudinal study design, it should be evaluated whether PCC, as an objective measure of sensory pathway integrity, provides additional information that allows separation between the good prognosis, poor prognosis, and crossover group.

## **Conclusion**

This study shows that subacute stroke subjects present PCC, indicating that afferent sensory information arrives at the cortex. Position-cortical coherence can even be detected in subjects with very poor motor function, who are unable to generate voluntary force. The presence of PCC on the sensorimotor area contralateral to the affected wrist is lower in poor function subjects compared with good function subjects. This shows that afferent pathways integrity and sensorimotor integration are affected in stroke survivors with poor motor function. In addition, in subjects with poor motor function, the lateralisation index showed that PCC is distributed more evenly between the lesioned and non-lesioned hemisphere during affected wrist perturbations than during non-affected wrist perturbations. Position-cortical coherence provides an objective measure of sensory pathway information transfer and processing after stroke and may provide additional insight into mechanisms of recovery of motor function after stroke.

## **Funding**

This study was financially supported by a fellowship granted to C.G.M. Meskers (F2011(1)-25) from the Dutch brain foundation (Hersenstichting), the Netherlands.

## REFERENCES

- 1 Kwakkel G, Kollen B, Lindeman E. Understanding the pattern of functional recovery after stroke: facts and theories. *Restor Neurol Neurosci* 2004; **22**: 281–299.
- 2 Connell LA, Lincoln NB, Radford KA. Somatosensory impairment after stroke: frequency of different deficits and their recovery. *Clin Rehabil* 2008; **22**: 758–767.
- 3 Schabrun SM, Hillier S. Evidence for the retraining of sensation after stroke: a systematic review. *Clin Rehabil* 2009; **23**: 27–39.
- 4 Baker SN. Oscillatory interactions between sensorimotor cortex and the periphery. *Curr Opin Neurobiol* 2007; **17**: 649–655.
- 5 Scott SH. Optimal feedback control and the neural basis of volitional motor control. *Nat Rev Neurosci* 2004; **5**: 532.
- 6 Williams ER, Soteropoulos DS, Baker SN. Coherence Between Motor Cortical Activity and Peripheral Discontinuities During Slow Finger Movements. *J Neurophysiol* 2009; **102**: 1296–1309.
- 7 Fries P. A mechanism for cognitive dynamics: neuronal communication through neuronal coherence. *Trends Cogn Sci* 2005; **9**: 474–480.
- 8 Varela F, Lachaux J-P, Rodriguez E, Martinerie J. The brainweb: Phase synchronization and large-scale integration. *Nat Rev Neurosci* 2001; **2**: 229.
- 9 Halliday DM, Conway B a, Farmer SF, Rosenberg JR. Using electroencephalography to study functional coupling between cortical activity and electromyograms during voluntary contractions in humans. *Neurosci Lett* 1998; **241**: 5–8.
- 10 Conway BA, Halliday DM, Farmer SF, Shahani U, Maas P, Weir AI et al. Synchronization between motor cortex and spinal motoneuronal pool during the performance of a maintained motor task in man. *J Physiol* 1995; **489**: 917–924.
- 11 Mima T, Steger J, Schulman AE, Gerloff C, Hallett M. Electroencephalographic measurement of motor cortex control of muscle activity in humans. *Clin Neurophysiol* 2000; **111**: 326–337.
- 12 Riddle CN, Baker SN. Manipulation of peripheral neural feedback loops alters human corticomuscular coherence. *J Physiol* 2005; **566**: 625–639.
- 13 Witham CL, Riddle CN, Baker MR, Baker SN. Contributions of descending and ascending pathways to corticomuscular coherence in humans. *J Physiol* 2011; **589**: 3789–3800.
- 14 Pohja M, Salenius S. Modulation of cortex-muscle oscillatory interaction by ischaemia-induced deafferentation. *Neuroreport* 2003; **14**: 321–324.
- 15 Mima T, Matsuoka T, Hallett M. Information flow from the sensorimotor cortex to muscle in humans. *Clin Neurophysiol* 2001; **112**: 122–126.
- 16 Campfens SF, Schouten AC, van Putten MJ, van der Kooij H. Quantifying connectivity via efferent and afferent pathways in motor control using coherence measures and joint position perturbations. *Exp brain Res* 2013; **c**: 141–153.
- 17 Ushiyama J, Suzuki T, Masakado Y, Hase K, Kimura A, Liu M et al. Between-subject variance in the magnitude of corticomuscular coherence during tonic isometric contraction of the tibialis anterior muscle in healthy young adults. *J Neurophysiol* 2011; **106**: 1379–1388.
- 18 Mendez-Balbuena I, Huethe F, Schulte-Monting J, Leonhart R, Manjarrez E, Kristeva R. Corticomuscular coherence reflects interindividual differences in the state of the corticomuscular network during low-level static and dynamic forces. *Cereb Cortex* 2012; **22**: 628–638.
- 19 Herrmann CS. Human EEG responses to 1-100 Hz flicker: resonance phenomena in visual cortex and their potential correlation to cognitive phenomena. *Exp brain Res* 2001; **137**: 346–353.
- 20 Campfens SF, van der Kooij H, Schouten AC. Face to phase: pitfalls in time delay estimation from coherency phase. *J Comput Neurosci* 2014; **37**: 1–8.
- 21 Ward NS, Brown MM, Thompson AJ, Frackowiak RSJ. Neural correlates of outcome after stroke: a cross-sectional fMRI study. *Brain* 2003; **126**: 1430–1448.
- 22 Serrien DJ, Strens LHA, Cassidy MJ, Thompson AJ, Brown P. Functional significance of the ipsilateral hemisphere during movement of the affected hand after stroke. *Exp Neurol* 2004; **190**: 425–432.
- 23 Fugl-Meyer AR, Jääskö L, Leyman I, Olsson S, Steglind S. The post-stroke hemiplegic patient. 1. a method for evaluation of physical performance. *Scand J Rehabil Med* 1975; **7**: 13–31.

- 24 Stolk-Hornsveld F, Crow JL, Hendriks EP, Van Der Baan R, Harmeling-van Der Wel BC. The Erasmus MC modifications to the (revised) Nottingham Sensory Assessment: a reliable somato-sensory assessment measure for patients with intracranial disorders. *Clin Rehabil* 2006; 20: 160–172.
- 25 Oostenveld R, Praamstra P. The five percent electrode system for high-resolution EEG and ERP measurements. *Clin Neurophysiol* 2001; 112: 713–719.
- 26 Oostenveld R, Fries P, Maris E, Schoffelen J-M. FieldTrip: Open source software for advanced analysis of MEG, EEG, and invasive electrophysiological data. *Comput Intell Neurosci* 2011; 2011: 1–9.
- 27 Severens M, Nienhuis B, Desain P, Duysens J. Feasibility of measuring event related desynchronization with electroencephalography during walking. *Conf Proc IEEE Eng Med Biol Soc* 2012; 2012: 2764–2767.
- 28 Bortel R, Sovka P. Approximation of statistical distribution of magnitude squared coherence estimated with segment overlapping. *Signal Processing* 2007; 87: 1100–1117.
- 29 De Vos M, Riès S, Vanderperren K, Vanrumste B, Alario F-X, Van Huffel S et al. Removal of muscle artifacts from EEG recordings of spoken language production. *Neuroinformatics* 2010; 8: 135–150.
- 30 Lincoln NB, Crow JL, Jackson JM, Waters GR, Adams SA, Hodgson P. The unreliability of sensory assessments. *Clin Rehabil* 1991; 5: 273–282.
- 31 Dukelow SP, Herter TM, Moore KD, Demers MJ, Glasgow JI, Bagg SD et al. Quantitative assessment of limb position sense following stroke. *Neurorehabil Neural Repair* 2010; 24: 178–187.
- 32 Kwakkel G, Kollen BJ, van der Grond J, Prevo AJH. Probability of regaining dexterity in the flaccid upper limb: impact of severity of paresis and time since onset in acute stroke. *Stroke* 2003; 34: 2181–2186.
- 33 Dobkin BH. Rehabilitation after Stroke. *N Engl J Med* 2005; 352: 1677–1684.
- 34 Nijland RHM, van Wegen EEH, Harmeling-van der Wel BC, Kwakkel G. Presence of finger extension and shoulder abduction within 72 hours after stroke predicts functional recovery: early prediction of functional outcome after stroke: the EPOS cohort study. *Stroke* 2010; 41: 745–750.









# Chapter 5

Position-cortical  
coherence as a marker  
of afferent pathway  
integrity early  
post-stroke, a  
prospective cohort study

Sarah B. Zandvliet  
Erwin E.H. van Wegen  
S. Floor Campfens  
Herman van der Kooij  
Gert Kwakkel  
Carel G.M. Meskers

*Neurorehabilitation and Neural Repair* 2020;  
34(4): 344–359



## ABSTRACT

**Background:** Addressing the role of somatosensory impairment, i.e. afferent pathway integrity, in post-stroke motor recovery may require neurophysiological assessment.

**Objective:** We investigated the longitudinal construct validity of position-cortical coherence (PCC), i.e. the agreement between mechanically evoked wrist perturbations and EEG, as a measure of afferent pathway integrity.

**Methods:** PCC was measured serially in 48 patients after a first-ever ischemic stroke in addition to Fugl-Meyer motor assessment of the upper extremity (FM-UE) and Erasmus modification of the Nottingham sensory assessment hand-finger sub-scores (EmNSA-HF, within 3 and at 5, 12 and 26 weeks post-stroke. Changes in PCC over time, represented by percentage presence of PCC (%PCC), mean amplitude of PCC over the affected (Amp-A) and non-affected hemisphere (Amp-N) and a lateralization index (L-index), were analyzed, as well as their association with FM-UE and EmNSA-HF. Patients were retrospectively categorized based on FM-UE score at baseline and 26 weeks post-stroke into high-and low-baseline recoverers and non-recoverers.

**Results:** %PCC increased from baseline to twelve weeks post-stroke ( $\beta$ :1.6%, CI:0.32–2.86%,  $P=0.01$ ), which was no longer significant after adjusting for EmNSA-HF and FM-UE. A significant positive association was found between %PCC, Amp-A and EmNSA-HF. Low-baseline recoverers ( $N=8$ ) showed longitudinally significantly higher %PCC than high-baseline recoverers ( $N=23$ ).

**Conclusions:** We demonstrated the longitudinal construct validity of %PCC and Amp-A as a measure of afferent pathway integrity. A high %PCC in low-baseline recoverers suggests that this measure also contains information on cortical excitability. Use of PCC as an EEG-based measure to address the role of somatosensory integrity to motor recovery post-stroke requires further attention.

---

## INTRODUCTION

Progress of time as a reflection of underlying spontaneous neurobiological recovery appears to account for 80–90% of the observed improvement in motor function and upper limb capacity in patients after stroke<sup>1–3</sup>. This accounts for approximately 70% of patients in which some patients are expected to recover, i.e. patients with a relatively high baseline motor function, while other patients are not, i.e. patients with a low baseline motor function. It is unknown why 20–30% of patients, the so-called non-recoverers, with a low motor function at 26 weeks post-stroke, fail to show spontaneous recovery<sup>4</sup>.

Patients with somatosensory impairments have a lower probability of regaining upper limb capacity than patients in whom this function is not affected<sup>3</sup>. Somatosensory impairments may influence motor recovery due to a tight interaction of the afferent and efferent pathways with a supposed cortical loop<sup>5,6</sup>. Disturbance of afferent pathway integrity is therefore important to study to understand motor recovery<sup>7</sup>. The Erasmus modification of the Nottingham sensory assessment of the upper extremity (EmNSA-UE) is a common and reliable clinical measure of somatosensory impairment in patients with stroke<sup>8</sup>. However, it is not a direct measure of afferent pathway integrity, and is not very responsive to change<sup>8–10</sup>.

Finger stimulation evoked somatosensory potentials could theoretically be an approach to study somatosensory processing, however may not be recommended due to the lack in reliability<sup>11</sup>. Cortical rebound responses in the beta band of the affected hemisphere measured with magneto-encephalography during manual passive finger movements, were found to correlate with the initial severity and recovery of motor activity as measured with the box and block test at 1 and 12 months post-stroke<sup>12</sup>. Joint position perturbations act as an external excitation signal for the proprioceptive system, primarily the Golgi tendon organs and muscle spindles, providing an interesting approach to study sensorimotor processing in severely affected patients<sup>12</sup>. Coherence between mechanical perturbations and subsequent cortical responses as measured with EEG, that is, position-cortical coherence (PCC), represent the unidirectional information transfer across the afferent pathways<sup>13,14</sup>. A cross-sectional study in stroke patients found significant differences in the presence of PCC between groups with poor and good motor function as measured by FM-UE using this system identification approach<sup>14</sup>. A similar protocol was used by Vlaar et al.<sup>15</sup>, who reported a reduced amplitude in ipsi-lesional cortical responses, quantified by a signal-to-noise ratio, in patients with severe somatosensory impairment (reduced EnMSA-UE score for more than two subtests) in patients in the chronic phase after stroke.

To address the potential added value of neurophysiological measures, prospective studies are required with fixed moments of measurements post-stroke to establish their construct

validity<sup>16</sup>. These neurophysiological biomarkers could help to improve the prediction of outcome, and enhance accurate selection of patients for clinical trials<sup>17,18</sup>.

We aimed to evaluate the longitudinal construct validity of PCC as a measure of afferent pathway integrity and its relation to post-stroke recovery, in a prospective cohort study with repetitive measurements at fixed time points within 3 and at 5, 12 and 26 weeks in patients after a first-ever ischemic stroke. We addressed the following research questions and corresponding hypotheses:

- (1) How does PCC change over the first 26 weeks post-stroke? We expected measures of PCC to show a significant change over time as a reflection of spontaneous neurobiological recovery. Due to these spontaneous neurobiological processes, we also expected this association over time to be influenced by the recovery of motor function, reflected by FM-UE and somatosensory function, reflected by EmNSA hand and finger subset (EmNSA-HF).
- (2) Does PCC relate to clinical somatosensory scores, and how does this compare with motor recovery? We expected measures of PCC to represent somatosensory integrity, i.e. to show a significant association with EmNSA-HF and not with FM-UE score. We hypothesized that these associations would be independent of time.
- (3) How does PCC relate to motor recovery post-stroke? We expected a higher PCC in recoverers compared to non-recoverers.

## METHODS

### Participants & design

The present cohort consisted of 48 patients with a first-ever ischemic stroke who were recruited within 3 weeks after stroke onset, between August 2012 and July 2016. A flowchart is displayed in Figure 5.1. Inclusion criteria were: (1) first-ever ischemic lesion within 3 weeks after onset, established with CT, MRI or clinically; (2) hemiparesis of the upper limb, i.e. a national institutes of health stroke scale, motor arm score of 1–4 points at the moment of inclusion; (3) no orthopedic limitations of the upper extremity; (4) no other neurological condition; (5) aged 18 years or older; (6) able to sit for 30 minutes with support; (7) no severe cognitive deficits (mini mental state examination score  $\geq 19$ ); and (8) sufficient motivation to participate. Measurements were performed at baseline, i.e. within 3 weeks, and repeated at 5, 12 and 26 weeks post-stroke. All procedures were in accordance with the declaration of Helsinki and were approved by the medical ethics

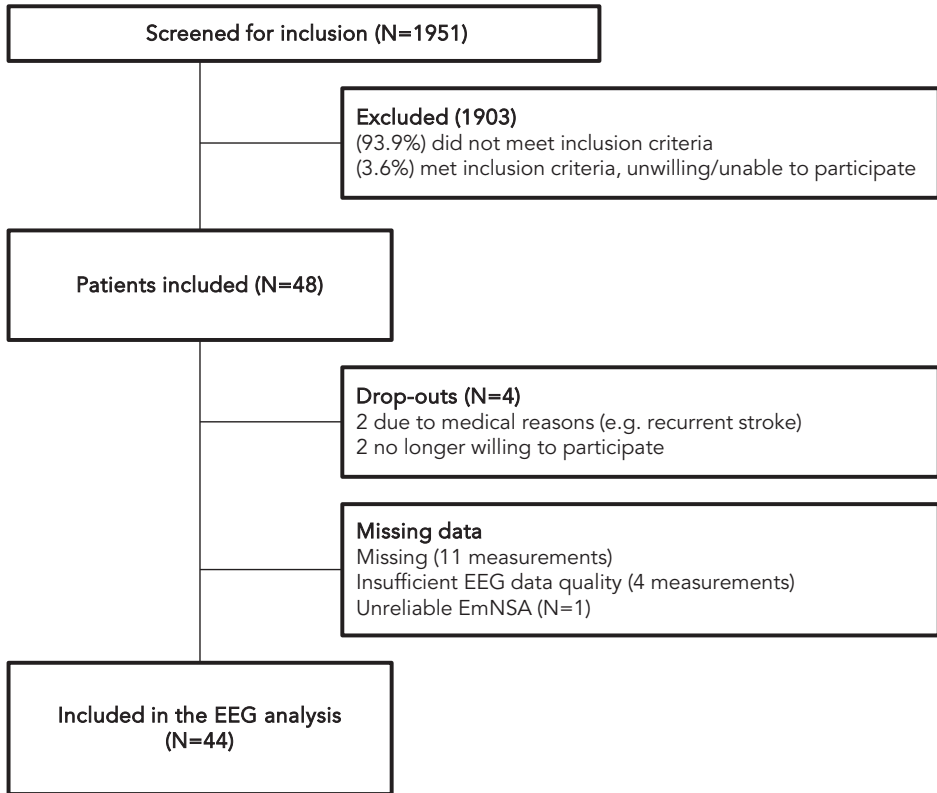


Figure 5.1 | Flow diagram.  
Inclusion flow diagram, number of patients (N).

committees of Leiden university medical center (NL39323.058.12, for eleven patients measured) and VU university medical center (NL 47079 029 14, for 37 patients measured). All participants gave their written informed consent.

### *Experimental set-up*

#### Measurement van

Eleven patients were measured in a hospital-based set-up at Leiden university medical center; the remaining 37 patients, from different stroke units in the Netherlands, were measured in a customized measurement van (Volkswagen Crafter, Wolfsburg, Germany, Figure 5.2a), certified as a medical room complying with NEN1010:2007 +C1:2008 +C1/A1 regulations of the VU university medical center. This measurement van enabled us to measure patients at their current site of residence, minimizing the burden of traveling.

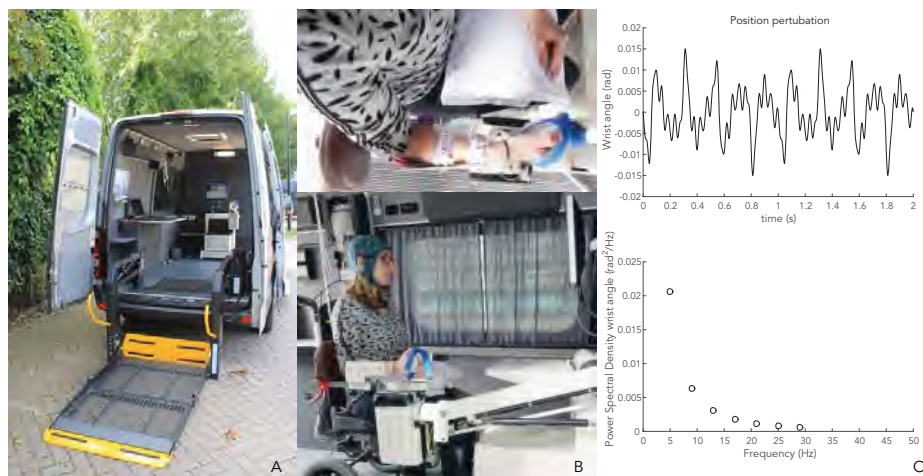


Figure 5.2 | Experimental setup.

Panel A: Experimental setup in the measurement van. Panel B: The patient's arm is placed in an arm rest and the wrist aligned with the axis of rotation of the wrist manipulator; the hand and arm are held in place with Velcro straps. Panel C: Two-second segment of the position perturbation. The position perturbation is a sum of sinusoids with a decreasing value of the power with frequency.

### Mechanical joint position perturbations

Patients were seated with their affected arm into a wrist manipulator (Wristlyzer, Moog Inc., Nieuw-Vennep, the Netherlands), a one-degree of freedom actuated rotating device by which angular perturbations can be imposed in both flexion and extension direction onto the wrist (Figure 5.2b).

### EEG equipment & signal acquisition

A 64-channel EEG set up with a 2048 Hz sample frequency was used to record cortical activity (Leiden university medical center: Refa, ANTneuro, Enschede, the Netherlands, measurement van: Refa, TMSi International, Oldenzaal, the Netherlands). Electrodes were placed on the skull using a 64-channel actively shielded EEG cap according to the international 10–20 system (TMSi, Oldenzaal, the Netherlands). Bipolar pairs of Ag/AgCl electrodes were used to monitor activity of the wrist muscles (Refa, TMSi International, Oldenzaal, the Netherlands). The force and position signals from the manipulator were recorded via optical isolation onto the same amplifier.

### Preparation

An EEG cap was placed over the head of the subject, after which the electrodes were filled with conductive gel. EMG electrodes were placed over the muscle bellies of the

m. flexor carpi radialis and the m. extensor carpi radialis longus of both the affected and the non-affected arm. Before placement, the skin was shaved, scrubbed and cleaned with alcohol to ensure a good signal-to-noise ratio. The height of the manipulator was adjusted to enable the subject to sit with their affected lower arm in 90 degrees elbow flexion and approximately 45 degrees shoulder abduction. The axis of rotation of the wrist manipulator was aligned with the axis of rotation of the affected wrist.

### ***Measurement protocol***

#### Relax task with multisine position perturbations

Subjects were asked to remain relaxed (monitored by EMG activity) while a series of perturbations were imposed onto the handle. The trials lasted 300 seconds and were performed 5 to 8 times. The wrist manipulator moved the affected wrist according to an unpredictable, smooth, periodic pattern consisting of a sum of sinusoids with power at frequencies of 5, 9, 13, 17, 21, 25 and 29 Hz with a fixed peak to peak amplitude of 0.03 rad (Figure 5.2c). More details about the used perturbation signal can be found in Campfens et al. 2013<sup>13</sup>.

#### Data processing

All EEG data processing was performed in Matlab 2012b (The MathWorks, Inc. Natick, Mass. USA) using the Fieldtrip toolbox for EEG analysis<sup>19</sup>. Statistical comparisons were performed using IBM SPSS statistics (v22) (IBM corporation, Armonk, NY, USA). MLwiN (2.28) was used for the mixed model analysis (center for multilevel modelling, University of Bristol, Bristol, UK).

#### EEG signal pre-processing

EEG signals were filtered with a high pass second order Butterworth filter with a cut-off frequency of 1 Hz to remove the linear trend and inspected for large artifacts, which were removed from the data. Subsequently the signals were band stop filtered between 49 and 51 Hz and its multiples with a second order Butterworth filter in both directions. Channels with no or very poor data quality were interpolated using a weighted method. An independent component analysis was applied to remove eye blink components based on the signal characteristic and topography. Components with a median frequency  $\geq 60$  Hz were considered EMG artifacts and removed. Signals were then manually inspected for remaining artifacts, which were removed from the data, and were divided into epochs of one second with 75% overlap.

PCC

The EEG segments were multiplied with a Hamming window and transformed to the frequency domain by a fast Fourier transformation. The power spectral density (*PSD*,  $\phi_{xx}(f)$ ) and cross spectral density (*CSD*,  $\phi_{xy}(f)$ ) were estimated.

$$\phi_{xx}(f) = \frac{1}{N} \sum_{i=1}^N X_i^*(f) \cdot X_i(f) \quad (1)$$

$$\phi_{xy}(f) = \frac{1}{N} \sum_{i=1}^N X_i^*(f) \cdot Y_i(f) \quad (2)$$

Subsequently the (magnitude-squared) coherence ( $C_{xy}(f)$ ) was calculated:

$$C_{xy}(f) = \frac{|\phi_{xy}(f)|^2}{\phi_{xx}(f) \phi_{yy}(f)} \quad (3)$$

PCC was calculated as the coherence between the measured perturbation signal and each EEG channel at the perturbed frequencies. The confidence limit (CL) was determined using an approximation method for overlapping segments, in which  $\alpha$  was set at 0.01 to achieve a confidence level of 99%<sup>14,20</sup>. The number of frequencies where the PCC amplitude exceeded the 99% confidence interval per electrode was taken and summed across the electrodes overlapping the contralateral (affected) sensorimotor area which consisted of the electrodes: FC1, FC3, FC5, C1, C3, C5, CP1, CP3 and CP5 for the left sensorimotor area and its equivalents for the right hemisphere. This sum was expressed as a percentage of the total number of 63 frequency bins, representing the percentage presence of PCC (%PCC). The amplitude of the mean significant PCC was calculated over the sensorimotor area of the affected (Amp-A) and the non-affected hemisphere (Amp-N). A lateralization index (L-index) was calculated to evaluate if the PCC was more lateralized towards the affected hemisphere (L-index > 1) or towards the non-affected hemisphere (L-index < 1).

$$\text{L-index} = 1 + \log_{10}(\text{Amp-A}) - \log_{10}(\text{Amp-N}) \quad (4)$$

Clinical measurements & subgrouping of patients

Fugl-Meyer motor assessment of the upper extremity (FM-UE)<sup>21</sup> and the Erasmus modification of the Nottingham sensory assessment (EmNSA)<sup>8</sup> were used as measures of motor function and somatosensory function. Since the mechanical perturbation via the manipulator was applied to the wrist, only the test items for hand and finger somatosensory function were used (EmNSA-HF) (maximal score of 20 points) for comparisons.



Patients were classified based on the amount of spontaneous neurobiological recovery they showed over time. The proportional recovery model<sup>4,22</sup> defined as:  $0.7 \cdot (66 - \text{initial score FM-UE}) + 0.4$ <sup>22</sup>, was used to determine the expected amount of spontaneous neurobiological recovery over time. Based on this expected improvement, patients who showed and patients who failed to show spontaneous neurobiological recovery (recoverers and non-recoverers), were distinguished by means of a hierarchical cluster analysis using Mahalanobis distances<sup>4</sup>. The categorization into low- and high-baseline was made based on the FM-UE baseline score, with a cut-off of 18 points, which was found to characterize non-recoverers in the study by Winters et al.<sup>4</sup>.

Retrospectively, we distinguished three motor recovery subgroups: (1) patients with an initial score of 18 points or higher on the FM-UE, who were expected to and showed spontaneous recovery (high-baseline recoverers); (2) patients with an initial score less than 18 points on the FM-UE, who nevertheless showed spontaneous recovery (low-baseline recoverers); (3) patients with an initial score less than 18 points on the FM-UE, who fail to show spontaneous recovery (non-recoverers).

## Statistics

Normality was examined by inspecting the histograms, q-q plots and Z-scores for skewness and kurtosis of the data, or the residuals when appropriate. A natural log transformation was applied when these assumptions were not met. If this transformation was not sufficient, or in the case of ordinal or nominal data, a non-parametric test was used. The significance level  $\alpha$  was set two-tailed at 0.05.

To examine the longitudinal change in the four PCC parameters over time, a mixed-model analysis was performed. EmNSA-HF and FM-UE were added to the model as a second step. A 10% change in effect estimates ( $\beta$ -values) was considered an improvement to the model<sup>23</sup>.

The longitudinal association of the four PCC parameters with EmNSA and FM-UE was examined using a second mixed-model analysis. We tested for possible interaction effects between measurement time point and EmNSA-HF as well as between time point and FM-UE.

A third association model was used to examine the relation between the PCC parameters and the different motor recovery subgroups, i.e. high- and low-baseline recoverers and non-recoverers. EmNSA-HF and measurement time points were tested for possible confounding and interaction effects.

In the fourth model, EmNSA-HF was taken as an outcome variable to examine the fixed effects between motor recovery subgroups over time and between subjects.

Differences in characteristics and baseline parameters between motor recovery subgroups were tested with a one-way ANOVA model in the case of continuous normally distributed data, with a Bonferroni correction for post-hoc analysis. The Kruskal Wallis test was used for ordinal data, with a Mann Whitney-U test for post-hoc analysis. Categorical/nominal variables were tested with Pearson's Chi squared, a 2x2 cross table was used for post-hoc analysis.

## RESULTS

48 of the 1951 screened patients were included for this prospective cohort study. During follow-up, 4 patients withdrew from the study. Among the 44 remaining patients, 165 measurements were performed. One EEG measurement was missing in 11 patients with additional missing of clinical measurement 2 of these patients. See Figure 5.1 for a flowchart. EEG data quality was not sufficient to calculate the parameters for 3 measurements in 1 patient and for 1 measurement in 1 other. In 17 of the 165 measurements 1 channel, and in 5 measurements, 2 channels were interpolated in the affected sensorimotor area from which PCC was calculated. This concerned 13 different patients. In 3 patients the same channel was interpolated in 2 measurements. An example of the measured signals and their quality is displayed in Supplementary Figure S5.1. In 1 patient, EmNSA-HF could not be measured correctly due to failure to understand the instructions. Baseline characteristics of the study population are displayed in Table 5.1.

### Changes in PCC over time

The association model for %PCC (Table 5.2a) revealed a significantly higher %PCC at the 12-week measurement time point; mean (M):97.4%, standard deviation (SD):2.7%, as compared to baseline, M:95.8%, SD:3.7%, 95% confidence interval (CI):0.32 to 2.86,  $P=0.01$ . These differences were no longer significant when the model was longitudinally corrected for EmNSA-HF and FM-UE.

No significant differences were found between the baseline and the other measurement time points, or between the 12- and 26-week measurements; M:96.9%, SD:2.7,  $\beta$ :-0.50, CI:-1.77 to 0.77,  $P=0.44$ . Amp-A, Amp-N and L-index did not differ significantly between measurement time points (see Table 5.2a and Supplementary Figure S5.2).

**Table 5.1** | Subject characteristics at baseline

Number of patients analysed	44
Time between stroke and baseline measurement (days) <sup>a</sup>	13.4 (5.3)
Age (y) <sup>a</sup>	64.5 (11.9)
Weight (kg) <sup>a</sup>	82 (17.2)
Height (m) <sup>a</sup>	1.74 (0.1)
Gender, male/female (N) <sup>b</sup>	28/16
Affected hemisphere, right/left (N) <sup>b</sup>	26/18
Bamford classification, LACI/PACI/TACI (N) <sup>b</sup>	24/16/4
Lesion location, cortical/subcortical/unknown (N) <sup>b</sup>	36/6/2
CIRS	4 (2–6)
NIHSS	5.5 (3–8)
FM-UE at baseline	20.5 (7–2.75)
FM-UE at 6 months ps	54.5 (18.5–62.75)
ARAT at baseline	3 (0–27.75)
ARAT at 6 months ps	50 (3–57)
EmNSA-hand and finger at baseline	18 (16–20)
EmNSA-hand and finger at 6 months ps	20 (19–20)
MI-UE at baseline	39 (9–65)
MI-UE at 6 months ps	53 (28–80)
MI-LE at baseline	76 (39–89.25)
MI-LE at 6 months ps	80 (58–100)

Characteristics and baseline values of all 44 patients. Unless mentioned otherwise, median and interquartile range are listed for each variable. a=continuous variable, mean and standard deviation are listed. b=categorical/nominal variable, number of patients is listed.

Abbreviations: years (y), kilogram (kg), meter (m), lacunar anterior circulation infarction (LACI), partial anterior circulation infarct (PACI), total anterior circulation infarct (TACI), cumulative illness rating scale (CIRS), national institute of health stroke scale (NIHSS), Fugl-Meyer motor assessment of the upper extremity (FM-UE), action research arm test (ARAT), Erasmus modification of the Nottingham sensory assessment (EmNSA), motricity index (MI), lower extremity (LE), upper extremity (UE), baseline is the first measurement of each subject within 3 weeks post-stroke, post-stroke (ps).

### Association of PCC with EmNSA-HF and FM-UE

A significant positive association was found between %PCC and EmNSA-HF;  $\beta$ :0.14, CI:0.02-0.26,  $P=0.02$ . The fixed effect estimate beta for EmNSA-HF changed by more than 10%, to  $\beta$ :0.12, CI:0.00-0.25,  $P=0.047$ , when measurement time was added to the model (see Table 5.2b). No interaction effects were found between measurement time points and EmNSA-HF (Figure 5.4).

A significant positive association was also found between Amp-A and EmNSA-HF;  $\beta$ :1.02 (ratio due to log transformation), CI:1.00-1.03,  $P=0.01$ . Including measurement time as a covariate did not change this model. No interaction effects were found between measurement time points and EmNSA-HF (Figure 5.4).

Table 5.2a | Results of the mixed-model analysis, development of the PCC parameters over time

Descriptives, measurement time points						
	Baseline	W5	W12	W26		
<b>%PCC</b>						
M, SD	95.82 ± 3.67	96.70 ± 3.40	97.39 ± 2.71	96.90 ± 2.65		
<b>Amp-A</b>						
Med, IQR	0.11 [0.09–0.13]	0.11 [0.10–0.13]	0.12 [0.10–0.16]	0.12 [0.10–0.16]		
<b>Amp-N</b>						
Med, IQR	0.11 [0.09–0.15]	0.12 [0.10–0.15]	0.13 [0.09–0.17]	0.13 [0.10–0.16]		
<b>L-index</b>						
M, SD	0.98 ± 0.12	0.96 ± 0.14	0.99 ± 0.14	0.98 ± 0.14		
<b>1</b>	Association model of PCC measures and time					
	Baseline	Base to W5	Base to W12	Base to W26		
<b>%PCC</b>						
β, CI	95.79 [94.85–96.73]	0.90 [-0.42–2.21]	1.56 [0.32–2.86]	1.09 [-0.19–2.37]		
P	-	0.86	0.01	0.10		
<b>Amp-A*</b>						
β, CI	0.11 [0.10–0.13]	0.99 [0.87–1.13]	1.10 [0.96–1.24]	1.09 [0.96–1.24]		
P	-	0.86	0.16	0.20		
<b>Amp-N*</b>						
β, CI	0.12 [0.11–0.13]	1.05 [0.91–1.21]	1.08 [0.94–1.24]	1.09 [0.94–1.25]		
P	-	0.51	0.27	0.25		
<b>L-index</b>						
β, CI	0.98 [0.94–1.03]	-0.02 [-0.08–0.04]	0.01 [-0.05–0.06]	0.00 [-0.06–0.06]		
P	-	0.42	0.86	1.00		

2 Association model of PCC measures and time, corrected for EmNSA-HF and FM-UE							
	Baseline	EmNSA-HF	FM-UE	Base to W5	Base to W12	Base to W26	
<b>%PCC</b>							
$\beta$ , CI	93.89 [91.71–96.07]	0.15 [0.03–0.28]	-0.02 [-0.04–0.01]	0.77 [-0.59–2.14]	1.22 [-0.13–2.58]	0.87 [-0.49–2.23]	
P	-	0.02	0.17	0.27	0.08	0.21	
<b>Amp-A*</b>							
$\beta$ , CI	0.09 [0.07–0.11]	1.02 [1.01–1.03]	1.00 [1.00–1.00]	0.98 [0.86–1.12]	1.05 [0.93–1.20]	1.06 [0.93–1.21]	
P	-	0.01	0.32	0.78	0.43	0.38	
<b>Amp-N*</b>							
$\beta$ , CI	0.1 [0.08–0.13]	1.01 [1.00–1.02]	1.00 [1.00–1.00]	1.03 [0.89–1.19]	1.05 [0.91–1.21]	1.05 [0.90–1.21]	
P	-	0.20	0.32	0.71	0.53	0.58	
<b>L-index</b>							
$\beta$ , CI	0.95 [0.85–1.04]	0 [0.00–0.01]	0 [0.00–0.00]	-0.02 [-0.08–0.04]	0 [-0.06–0.06]	0.01 [-0.05–0.07]	
P	-	0.32	0.32	0.52	0.95	0.80	

Association models with percentage of presence of position-cortical coherence (% PCC), mean PCC amplitude for the affected hemisphere (Amp-A), mean amplitude for the non-affected hemisphere (Amp-N) and lateralisation index (L-Index) as dependent variables.

Results of the mixed-model analysis: 1) Development of the PPC parameters over time. 2) The models corrected for EmNSA-HF and FM-UE.

\* Indicates a ratio due to log-transformation.

Abbreviations: Fugl-Meyer motor assessment of the upper extremity (FM-UE), Erasmus modification of the Nottingham sensory assessment (EmNSA), hand and finger subset (HF), motricity index (MI), lower extremity (LE), upper extremity (UE), mean (M), standard deviation (SD), median (med), inter quartile range (IQR), 95% confidence interval (CI), probability value for the tested model (P). Baseline is the first measurement of each subject within 3 weeks post-stroke, week post-stroke (W).

Table 5.2b | Results of the mixed-model analysis, association of the PCC parameters with FM-UE and EmNSA-HF

1	FM-UE		FM-UE corrected for time				
	Constant	FM-UE	Baseline	FM-UE	Base to W5	Base to W12	Base to W26
%PCC	$\beta$ , CI	0.00 [-0.02-0.02]	96.00 [94.90-97.11]	-0.01 [-0.03-0.02]	0.95 [-0.44-2.34]	1.59 [0.23-2.95]	1.20 [-0.19-2.59]
	P	0.93	-	0.52	0.18	0.02	0.09
	Amp-A*						
%PCC	$\beta$ , CI	1.00 [1.00-1.00]	0.11 [0.01-0.13]	1.00 [1.00-1.00]	0.99 [0.87-1.13]	1.08 [0.95-1.23]	1.09 [0.95-1.24]
	P	1.00	-	1.00	0.86	0.26	0.22
	Amp-N*						
%PCC	$\beta$ , CI	1.00 [1.00-1.00]	0.11 [1.00-1.00]	1.00 [1.00-1.00]	1.03 [0.89-1.20]	1.06 [0.92-1.22]	1.06 [0.92-1.22]
	P	0.046	-	0.046	0.66	0.45	0.44
	L-index						
%PCC	$\beta$ , CI	0.00 [0.00-0.00]	1.00 [0.95-1.05]	0.00 [0.00-0.00]	-0.02 [-0.08-0.04]	0.01 [-0.05-0.07]	0.01 [-0.05-0.07]
	P	-	-	1.00	0.51	0.76	0.71

2		EmNSA-HF corrected for time						
EmNSA-HF		EmNSA-HF	Baseline	EmNSA-HF	Base to W5	Base to W12	Base to W26	
<b>%PCC</b>								
$\beta$ , CI	94.21 [92.03-96.39]	0.14 [0.02-0.26]	93.93 [91.7-96.15]	0.12 [0.00-0.25]	0.67 [-0.64-1.98]	1.10 [-0.23-2.43]	0.68 [-0.64-2.00]	
P	-	0.02	-	0.047	0.31	0.11	0.31	
<b>Amp-A*</b>								
$\beta$ , CI	0.09 [0.07-0.11]	1.02 [1.00-1.03]	0.09 [0.07-0.11]	1.02 [1.00-1.03]	0.98 [0.86-1.11]	1.04 [0.92-1.19]	1.05 [0.93-1.19]	
P	-	0.01	-	0.01	0.72	0.51	0.46	
<b>Amp-N*</b>								
$\beta$ , CI	0.10 [0.08-0.13]	1.01 [1.00-1.02]	0.01 [0.08-0.13]	1.01 [1.00-1.03]	1.04 [0.89-1.20]	1.06 [0.92-1.22]	1.06 [0.92-1.22]	
P	-	0.046	-	0.12	0.64	0.45	0.46	
<b>L-index</b>								
$\beta$ , CI	0.94 [0.85-1.04]	0.00 [0.00-0.01]	0.95 [0.85-1.05]	-0.03 [-0.09-0.04]	-0.01 [-0.07-0.06]	-0.00 [-0.06-0.06]	0.00 [0.00-0.01]	
P	-	0.51	-	0.4	0.87	0.92	0.51	

Association models with percentage of presence of position-cortical coherence (% PCC), mean PCC amplitude for the affected hemisphere (Amp-A), mean amplitude for the non-affected hemisphere (Amp-N) and lateralisation index (l-Index) as dependent variables.

Results of the mixed-model analysis: 1) Association of the PCC parameters with FM-UE and EmNSA-HF; 2) The models after correction for time.

\* Indicates a ratio due to log-transformation.

Abbreviations: Fugl-Meyer motor assessment of the upper extremity (FM-UE); Erasmus modification of the Nottingham sensory assessment (EmNSA), inter quartile range (IQR), 95% confidence interval (CI), probability value for the tested model (P). Baseline is the first measurement of each subject within 3 weeks post-stroke, week post-stroke (W).



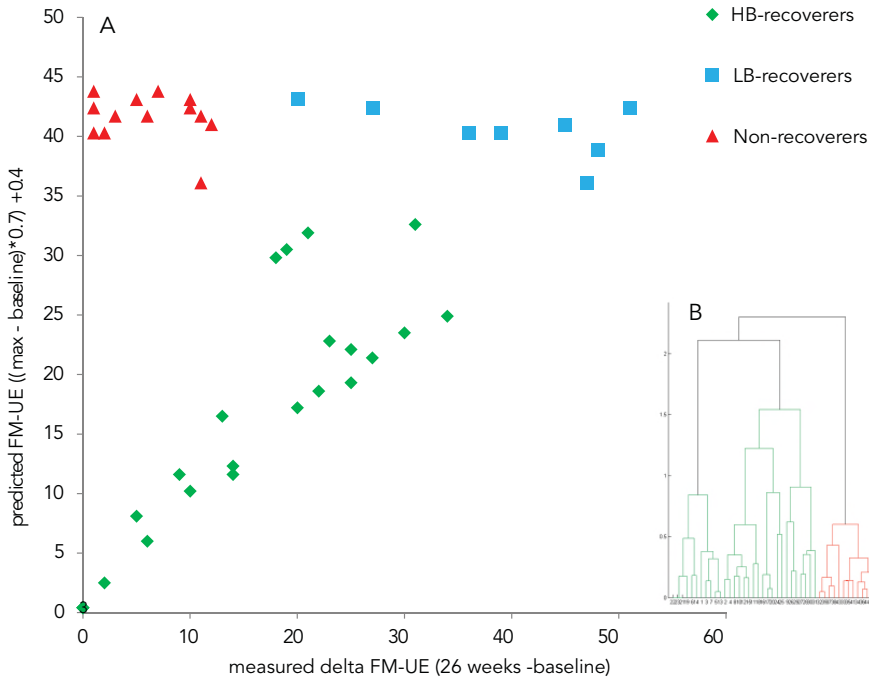
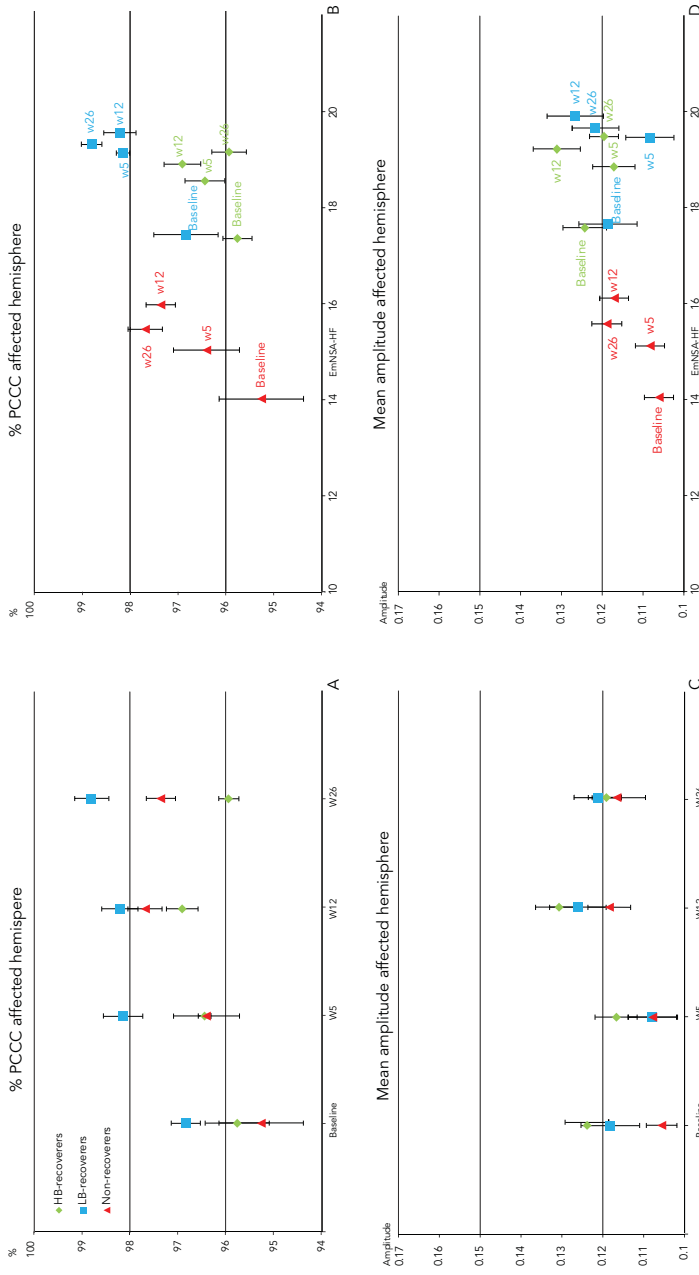


Figure 5.3 | Cluster analysis to distinguish recovery groups.

The proportional recovery model displaying the measured improvement on the Fugl-Meyer motor assessment of the upper extremity (FM-UE) relative to the predicted improvement of FM-UE at 6 months post-stroke =  $0.7 \times (66 - \text{FM-UE-baseline}) + 0.4$ . A cluster analysis was used to distinguish between recoverers and non-recoverers. Subjects with a baseline score  $\geq 18$  points on the FM-UE were classified as having a high-baseline (HB), (high-baseline recoverers in green), while subjects with  $< 18$  points on the FM-UE were classified as having a low-baseline (LB). Eight of these patients were classified as recoverers (low-baseline recoverers in blue). Thirteen patients were classified as non-recoverers (in red). In case overlapping data points, the numbers are indicated. The B panel shows the hierarchical cluster analysis in which green indicates the fitters and red the non-fitters to the proportional recovery model.

%PCC and Amp-A were not associated with FM-UE. While Amp-N and FM-UE were significantly associated;  $\beta: 1.00$  (ratio due to log transformation), CI: 1.00–1.00,  $P=0.046$ . This association remained significant when measurement time was added to the model. The positive association found between Amp-N and EmNSA-HF;  $\beta: 1.01$  (ratio due to log transformation), CI: 1.00–1.02,  $P=0.046$ , did not change when corrected for measurement time. No interaction effects were found.

No association was found between L-index and FM-UE or EmNSA-HF, (Table 5.2b and Supplementary Figures S5.3 and S5.4).



**Figure 5.4** | Percentage of significant position-cortical coherence (%PCC) and amplitude of PCC in the affected hemisphere in the recovery groups, over time and in relation with EmNSA-HF. Panel B: % PCC versus the EmNSA-HF. Panel C: Mean amplitude of PCC of all electrodes overlapping the sensorimotor cortex of the affected hemisphere over time. Panel D: Amplitude of PCC versus the EmNSA-HF. Position-cortical coherence (PCC) calculated for the electrodes overlapping the sensorimotor cortex for high-baseline recoverers (HB-recoverers), low-baseline recoverers (LB-recoverers) and non-recoverers, at baseline and at 5, 12, and 26 weeks post-stroke. The Erasmus modification of the Nottingham sensory assessment of the hand and fingers (EmNSA-HF) was measured at the same time points. Error bars indicate the standard error of the mean. Subgroups were distinguished by means of a hierarchical cluster analysis as displayed in Figure 5.3.

Table 5.3 | Differences at baseline between recovery pattern subgroups, i.e. high-and low-baseline recoverers and non-recoverers

	Non-recoverers (NR)	Low-baseline recoverers (LB-R)	High-baseline recoverers (HB-R)	All groups	NR to LB-R	LB-R to HB-R	NR to HB-R
	N=13	N=8	N=23	P	P	P	P
Time between stroke and baseline measurement (days) <sup>a</sup>	14.85 (4.4)	11.25 (5.2)	13.39 (5.8)	.33	-	-	-
Age (y) <sup>a</sup>	65.46 (14.0)	61.88 (10.3)	64.96 (11.5)	.78	-	-	-
Gender, male/female (n) <sup>b</sup>	8/5	4/4	16/7	.60	-	-	-
Affected hemisphere, right/left (n) <sup>b</sup>	9/4	1/7	16/7	.01	.011	.005	-
Bamford class, LACI/PACI/TACI (n) <sup>b</sup>	6/4/3	5/3/0	13/9/1	.34	-	-	-
Lesion location, cortical/subcortical/unknown (n) <sup>b</sup>	12/1/0	6/0/2	18/5/0	.02	.133	.024	.277
CIRS	4 (2.5-6)	3 (2-5.75)	4 (3-7)	.64	-	-	-
NIHSS baseline	8 (7-13)	8.5 (6-10)	4 (2-5)	<.001	0.74	.001	.001
FM-UE at baseline	7 (5-8.5)	8.5 (6-10.5)	42 (33-55)	<.001	0.21	<.001	<.001
ARAT at baseline	0 (0-2)	0 (0-0)	24 (11-42)	<.001	0.38	<.001	<.001
EmNSA-hand and finger at baseline	16 (9-19)	18 (16.5-19.75)	18.5 (17.75-20)	.22	-	-	-
Mi-UE at baseline	0 (0-26.50)	10 (2.25-32.75)	65 (53-76)	<.001	0.24	<.001	<.001
Mi-LE at baseline	28 (4.50-39.5)	53 (31.5-68.5)	75 (53-91)	.001	0.049	.069	<.001

Recovery pattern subgroup classification is based on the proportional recovery model in which predicted improvement of FM-UE at 6 months post-stroke=0.7\*(66-FM-UE-baseline) +0.4. A cluster analysis was used to distinguish between recoverers and non-recoverers. Subjects with a baseline score ≥18 points were classified as having a high baseline (HB-R), while subjects with <18 points on the FM-UE were classified as having a low-baseline. Eight of these patients were classified as recoverers (LB-R). Thirteen patients were classified as non-recoverers (NR). Unless mentioned otherwise, median and interquartile ranges are given for each variable per group. A Kruskal-Wallis test was used to test for differences between the three groups, while a Mann Whitney-U test was used for post-hoc analysis. continuous variable (a), mean and standard deviation are listed. A one-way ANOVA model was used to test the differences between the three groups. Categorical/nominal variable (b): number of patients is listed. Pearson's Chi squared test was used to test for differences between the three circulation infarct groups, and a 2x2 cross table was used for post-hoc analysis. Abbreviations: years (y), number of subjects per group (N), lacunar anterior circulation infarction (LACI), Partial anterior circulation infarct (PACI), total anterior circulation infarct (TACI), cumulative illness rating scale (CIRS), national institute of health stroke scale (NIHSS), Fugl-Meyer motor assessment of the upper extremity (FM-UE), action research arm test (ARAT), Erasmus modification of the Nottingham sensory assessment (EmNSA), motricity index (MI), lower extremity (LE), upper extremity (UE), probability value for the tested model (P), baseline is the first measurement of each subject within 3 weeks post-stroke, post-stroke (ps).

## Subgroups of motor recovery patterns

Of the 44 patients, 31 showed spontaneous recovery and were classified as recoverers. Eight of these recoverers started with a FM-UE score lower than 18 and were categorized as low-baseline recoverers. The remaining 23 patients had a FM-UE score of 18 points or higher at baseline and were categorized as high-baseline recoverers. 13 patients were classified as non-recoverers, all of them having a FM-UE score lower than 18 points at baseline (Figure 5.3).

## Baseline characteristics of patient subgroups

Differences between high-baseline recoverers, low-baseline recoverers and non-recoverers are presented in Table 5.3. Non-recoverers and low-baseline recoverers significantly differed only on by their affected hemisphere; non-recoverers: 9 right- vs. 4 left-sided affected; low-baseline recoverers: 1 right- vs. 7 left-sided,  $P=0.01$ , and on the motricity index of the lower extremity; non-recoverers: median: 28, interquartile range (IQR):4.5–39.5, low-baseline recoverers: median: 53, IQR:31.5–68.5,  $P=0.05$  (Table 5.3).

## Association of EmNSA-HF with motor recovery subgroups

EmNSA-HF showed a significantly lower value in non-recoverers:  $M:15.37$ ,  $SD:6.24$  as compared to low-baseline recoverers:  $M:19.13$ ,  $SD:1.65$ ;  $\beta:-3.38$ ,  $CI:-4.73$  to  $-2.02$ ,  $P<0.001$  and compared to high-baseline recoverers  $M:18.75$ ,  $SD:2.59$ ;  $\beta:-3.76$ ,  $CI:-5.51$  to  $-2.01$ ,  $P<0.001$ .

## Association of PCC measures with motor recovery subgroups

A significantly higher %PCC was found in the low-baseline,  $M:98.0$ ,  $SD:2.2$ , as compared to the high-baseline recoverers:  $M:96.2$ ,  $SD:3.0$ ,  $\beta:1.75$   $CI:0.42-3.08$ ,  $P=0.01$ . A non-significantly lower %PCC was found in the non-recoverers:  $M:96.7$ ,  $SD:3.7$ , as compared to low-baseline recoverers;  $\beta:-1.25$ ,  $CI: -2.71-0.21$ ,  $P=0.09$ . No difference was found between high-baseline recoverers and non-recoverers. EmNSA-HF as a covariate improved the model, this correction attenuated the difference between low-baseline recoverers and non-recoverers (Table 5.4 and Figure 5.4).

A lower Amp-A was found in non-recoverers median: 0.11, IQR: 0.09–0.14, as compared to low-baseline recoverers, median: 0.14, IQR:0.1–0.2;  $\beta:0.86$  (ratio due to log transformation),  $CI: 0.76-0.99$ ,  $P=0.03$ . A non-significantly higher Amp-A was found in low-baseline recoverers as compared to high-baseline recoverers: median: 0.11, IQR:0.10–0.14;  $\beta:1.12$ ,  $CI:0.99-1.26$ ,  $P=0.09$ .

Table 5.4 | Results of the mixed-model analysis, association of the PCC parameters in the subgroups, i.e. high-and low-baseline recoverers and non-recoverers, over time and in relation with EmNSA-HF

Descriptives recovery pattern subgroups					
	HB-R	LB-R	NR		
%PCC					
M, SD	96.24 ± 3.01	97.99 ± 2.20	96.72 ± 3.71		
Amp-A					
Med, IQR	0.11 [0.10-0.14]	0.14 [0.11-0.16]	0.11 [0.09-0.14]		
Amp-N					
Med, IQR	0.13 [0.10-0.16]	0.11 [0.09-0.14]	0.11 [0.09-0.17]		
L-index					
M, SD	0.96 ± 0.14	1.05 ± 0.13	0.97 ± 0.13		
EmNSA-HF					
M, SD	18.75 ± 2.59	19.13 ± 1.65	15.37 ± 6.24		
1 Association model of PCC measures and subgroups					
	HB-R	LB-R	HB-R to LB-R	LB-R to NR	HB-R to NR
%PCC					
β, CI	96.23 [95.55-96.91]	97.98 [96.84-99.12]	1.75 [0.42-3.08]	-1.25 [-2.71-0.21]	0.50 [-0.64-1.64]
P	-	-	0.01	0.09	0.39
Amp-A*					
β, CI	0.12 [0.11-0.13]	0.13 [0.12-0.15]	1.12 [0.99-1.26]	0.86 [0.76-0.99]	0.97 [0.87-1.07]
P	-	-	0.08	0.03	0.51
Amp-N*					
β, CI	0.13 [0.12-0.14]	0.12 [0.11-0.13]	0.93 [0.81-1.06]	1.04 [0.89-1.20]	0.96 [0.86-1.08]
P	-	-	0.26	0.61	0.50

<b>L-index</b>						
$\beta$ , CI	0.97 [0.94–0.99]	1.05 [1.00–1.09]	0.08 [0.03–0.14]	-0.08 [-0.14–0.02]	0.00 [-0.05–0.05]	
P	-	-	0.004	0.01	0.97	
<b>EmNSA-HF</b>						
$\beta$ , CI	18.75 [17.92–19.57]	19.13 [17.75–20.51]	0.38 [-1.23–1.99]	-3.38 [-4.73–2.02]	-3.76 [-5.51–-2.01]	
P	-	-	0.64	<0.001	<0.001	
<b>2 Association model of PCC measures and subgroups, corrected for EmNSA-HF</b>						
	HB-R	LB-R	EmNSA-HF	HB-R to LB-R	LB-R to NR	HB-R to NR
<b>%PCC</b>						
$\beta$ , CI	93.57 [91.19–95.96]	95.16 [92.59–97.73]	0.15 [0.03–0.27]	1.59 [0.34–2.84]	-0.85 [-2.27–0.57]	0.74 [-0.39–1.87]
P	-	-	0.02	0.01	0.24	0.20
<b>Amp-A*</b>						
$\beta$ , CI	0.09 [0.07–0.11]	0.01 [0.08–0.13]	1.02 [1.00–1.03]	1.10 [0.98–1.24]	0.90 [0.79–1.03]	1.00 [0.89–1.11]
P	-	-	0.01	0.10	0.14	0.94
<b>Amp-N*</b>						
$\beta$ , CI	0.10 [0.08–0.13]	0.09 [0.07–0.12]	1.01 [1.00–1.03]	0.91 [0.80–1.04]	1.09 [0.93–1.27]	0.99 [0.88–1.12]
P	-	-	0.06	0.18	0.30	0.87
<b>L-index</b>						
$\beta$ , CI	0.95 [0.84–1.05]	1.03 [0.92–1.14]	0.00 [-0.01–0.01]	0.08 [0.03–0.14]	-0.08 [-0.14–0.02]	0.00 [-0.05–0.05]
P	-	-	0.74	0.003	0.01	0.97

Association model with percentage of presence of position-cortical coherence (% PCC), mean PCC amplitude for the affected hemisphere (Amp-A), mean amplitude for the non-affected hemisphere (Amp-N) and lateralisation index (L-Index) as dependent variables.

1) Association of the PCC parameters and EmNSA-HF with recovery subgroups. 2) Association with the PCC parameters after correction for EmNSA-HF.

Subjects were classified into three recovery subgroups: subjects with a high-baseline score  $\geq 18$  points on the FM-UE (HB-R), while subjects with  $< 18$  points on the FM-UE were classified as having a low-baseline. Eight of these patients were classified as recoverers (LB-R). Thirteen patients were classified as non-recoverers (NR). \* Indicates a ratio due to log-transformation.

Abbreviations: Fugl-Meyer motor assessment of the upper extremity (FM-UE), Erasmus modification of the Nottingham sensory assessment (EmNSA), mean (M), standard deviation (SD), median (med), inter quartile range (IQR), 95% confidence interval (CI), probability value for the tested model (P). Baseline is the first measurement of each subject within 3 weeks post-stroke, weeks post stroke (w).

No difference in Amp-A was found between high-baseline recoverers and non-recoverers. Measurement time as a covariate did not change the model by 10% or more (Table 5.4 and Figure 5.4). No difference in Amp-N was found between the subgroups.

A significantly lower L-index was found in non-recoverers: M:0.97, SD:0.13 as compared to low-baseline recoverers: M:1.05, SD:0.13  $\beta$ : -0.08, CI: -0.14 to -0.02,  $P=0.01$ , as well as among high-baseline recoverers: M:0.96, SD:0.14 as compared to low-baseline recoverers  $\beta$ :0.08, CI:0.03–0.14,  $P=0.003$ . Adding EmNSA-HF as a covariate did not influence these differences (Table 5.4 and Supplementary Figure S5.3).

## DISCUSSION

We conducted a longitudinal cohort study with repetitive measurements at fixed time points post-stroke combining EEG with clinical measures of sensorimotor function of the upper limb after stroke. We found a significant difference between percentage of position-cortical coherence (%PCC) at baseline and at 12 weeks post-stroke, a difference which attenuated after correction for level of somatosensory and motor impairment. No significant difference was found between the 12- and 26-week measurements or between the baseline and 26-week measurements. This time window of 12 weeks post-stroke is in line with mechanisms of spontaneous neurobiological recovery, which is predominant in the first 5 to 8 weeks post-stroke<sup>2,24</sup>. We therefore confirm our first hypothesis that %PCC changes over time as a reflection of spontaneous neurobiological recovery. This result also confirms the need for repetitive measurements to quantify the non-linear time-dependent dynamics of cortical markers in the recovery of upper limb function after stroke<sup>25</sup>. The significant positive longitudinal relation found between %PCC and EmNSA-HF, Amp-A and EmNSA-HF and between Amp-N and EmNSA-HF confirms the longitudinal construct validity of %PCC and Amp-A as a measure of afferent pathway integrity.

We found a significant association between motor function in terms of FM-UE score and Amp-N, while the other measures did not show this association. The present result is therefore not in line with an earlier cross-sectional study<sup>14</sup>, which found a significantly higher %PCC in the group of patients with FM-UE scores >18 points as compared to the more severely affected patients. The lower overall %PCC, ranging from 35 to 95%<sup>14</sup> compared to the 85 to 100% range found in our study may be explained by the differences in artifact removal, since we used independent component analysis and interpolated poor data channels.



We found a significant association between Amp-N and FM-UE, irrespective of the time of assessment post-stroke. Interestingly, we found no difference between motor recovery subgroups (high-baseline recoverers, low-baseline recoverers and non-recoverers). Although the L-index did not show an association with FM-UE nor with EmNSA-HF, the L-index did show significant more lateralization towards the affected hemisphere in the low-baseline recoverers, while the expected and non-recoverers showed lateralization towards the non-affected hemisphere. Unlike %PCC and Amp-A, these differences could not be explained by differences in EmNSA-HF. It is therefore unclear what exactly is represented by the L-index and Amp-N.

70.5% of the included patients in this study showed spontaneous neurobiological recovery including all patients with a FM-UE score of 18 points or higher at baseline, which is comparable to previously studies<sup>4,22</sup>. The longitudinal association between EmNSA-HF and the subgroups revealed a significantly higher EmNSA-HF in both low and high-baseline recoverers as compared to the non-recoverers, and no difference between the low and high-baseline recoverers.

We could not confirm our third hypothesis that PCC would be lower in non-recoverers compared to recoverers and would not differ between high- and low-baseline recoverers. The expected construct of %PCC, representing solely the integrity of afferent pathways may therefore be incomplete. A possible explanation for the lower values of %PCC in both the non-recoverers and the high-baseline recoverers could be that %PCC also reflects enhanced activity of cortical networks next to representing afferent pathway integrity. A decrease in beta-rebound, i.e. an increase in cortical excitability, in response to tactile finger stimulation and passive finger movement, has been previously linked to better functional outcome<sup>12,26</sup>. Possibly this increased cortical excitability may only be needed after stroke when motor function is severely affected, this compensatory mechanism might then be failing in the non-recoverers.

Parkkonen et al.<sup>12</sup> used passive finger movements to evoke cortical responses, a task comparable to that is used in our study, in a cohort of 23 patients who were measured within one week and at one month and one year post-stroke. In addition to a significant positive correlation between cortical excitability (decrease in beta-rebound measured with MEG) and functional outcome, a decrease in cortical excitability was found over time, which correlated with functional recovery<sup>12</sup>. It is possible that in our study, a normalization of cortical excitability had already occurred in the high-baseline recoverers before the first measurement within 3 weeks, reflected by a lower %PCC, paralleling their functional recovery.

Nicolo et al.<sup>27</sup> suggested that the association between coherence measures of functional connectivity and clinical improvement after stroke might reflect neurotransmitter changes, and that GABAergic processes in particular are reflected in the beta band. Our perturbation signal was largely within the beta band range, therefore PCC might reflect a similar process. Our study provides evidence that cortical excitability as well as afferent pathway integrity might be contained in %PCC. Establishing the direct link between EEG markers and synaptic processes requires further research that can bridge the gap between animal models and early post-stroke studies in humans<sup>28</sup>.

To the best of our knowledge this study is the largest prospectively conducted cohort study on EEG and upper limb function in stroke so far. The study incurred only a few drop outs and missing serial EEG-measurements. The high compliance may be attributable to the use of a measurement van allowing to collect high-quality data. No differences between the group measured in the hospital and the van for all four PCC parameters were found, and data quality was comparable. The measurement van could be a promising way to measure patients in their local community which reduces burden and costs. It may be a new way to explore the longitudinal relationship between derivatives of brain plasticity, such as EEG, and clinical somatosensory and motor recovery very early post-stroke. Importantly, differences in timing of assessments could be avoided as was recommended by the SRRR task force<sup>29</sup>.

### **Limitations and future directions**

Four different outcome parameters of PCC were calculated for this study and tested in separate mixed model analysis. No adjustment for multiple comparisons was made since most results were obtained from one model and the conclusions of the paper were not based on single significant result. Values displayed in Table 5.3 and 5.4 should be interpreted as such.

Our study included significantly more patients with a subcortical lesion in the high compared to the low-baseline recoverers, while no significant difference was found between the non-recoverers and recoverers. We recommend for future studies to make MRI-scans several months after stroke to provide detailed information with respect to the exact lesion volume and location, to explain individual differences in recovery. It could be informative to compare anatomical afferent pathway integrity by Diffusor Tensor Images with functional alternation measured with EEG<sup>15</sup>.

The presence of PCC and the Amp-A (PCC amplitude of the affected hemisphere) show generally the same pattern in the association models. The small range of 16% at baseline

(from 84 to 100%), with 6 out of the 44 patients at 100% PCC, suggest a ceiling effect of %PCC. %PCC indicates whether or not coherence is detected in specific electrodes and frequency bins. As such, it may be more a dichotomous than a continuous measure. In contrast, Amp-A is more sensitive to changes and not restricted by a ceiling effect, however it is more prone to noise than %PCC. Future studies are needed to improve the reliability of Amp-A by for example by converting the signals from an electrode to a source level. Analyzing Amp-A on a source level, if successful, would also resolve the multiple comparison problem that generally exists when analyses are performed on an electrode level.

PCC quantifies only the linear response while Vlaar et al. has shown that only about 20% of the cortical response can be found on the perturbed frequencies and can therefore be quantified as linear<sup>30</sup>. The perturbation signal was designed to be able to investigate the non-linear responses, yet a linear approach was the first choice to investigate longitudinal changes of neural pathways. Non-linear coherence measures may yield further information on the response to a wrist joint perturbation in the highly non-linear closed-loop afferent pathway system, which would be interesting to study in patients with a stroke<sup>31</sup>.

Other neurophysiological measures to study afferent pathway integrity post-stroke were proposed such as muscle stretch evoked potentials (StrEP)<sup>32</sup>. In a cross-sectional study, this particular marker was found to be consistent across conditions and sessions however was found not to differ significantly between patients with good and poor motor recovery<sup>14,32</sup>. While the ultimate marker for afferent pathway integrity has not yet been found, longitudinal studies are needed to evaluate possible markers in post-stroke recovery that have shown good reproducibility. Potential biomarkers for afferent pathway integrity, like PCC, StrEP, median nerve stimulation and other emerging methods, should be compared and evaluated in longitudinal studies.

Patients with an initially mild to moderate impairment, classified as a high baseline on FM-UE of 18 points or more, all complied with the proportional recovery model of Prabhakaran et al.<sup>22</sup>. Note that the variability in this group is likely to be larger than the proportional recovery model suggests<sup>33</sup>. To show the construct validity of the %PCC and Amp-A as a measure of afferent pathway integrity, also patients with a maximal score (13 out of 44 patients), were included in the current dataset.

Of the clinical assessments, only lower extremity function, expressed in the motricity index, was found to differ significantly between non-recoverers and low-baseline recoverers at baseline, while the EmNSA-HF did not differ between subgroups.

In order for %PCC or Amp-A to be of clinical use, it needs to be able to correctly predict individual recovery post-stroke, which is not yet feasible at this moment. More advanced analysis, such as the non-linear dynamics of the signal and source localization, need to be explored to better understand the biological meaning of PCC and differentiate between the information it contains on afferent pathway integrity and cortical excitability.

## Conclusions

EEG derived percentage of PCC (%PCC) showed a change over time in line with processes of spontaneous neurobiological recovery. We demonstrated the longitudinal construct validity of %PCC and Amp-A as measures of afferent pathway integrity.

However, a higher %PCC in low-baseline recoverers compared to non-recoverers and high-baseline recoverers suggests that this biomarker may also contain information on cortical excitability next to afferent pathway integrity. More efforts are needed to distinguish these processes before %PCC and Amp-A can be used as biomarkers for predicting post-stroke motor recovery.

## Acknowledgements

We would like to thank Aukje S. Andringa for her contribution in the organization of the study and collection of the data and Caroline Winters, Mique Saes, Dirk Hoevenaars, Luuk Haring and Elza van Duijnhoven for their contributions in the data collection. We also would like to thank Jos W. Twisk for his help with the statistical analysis.

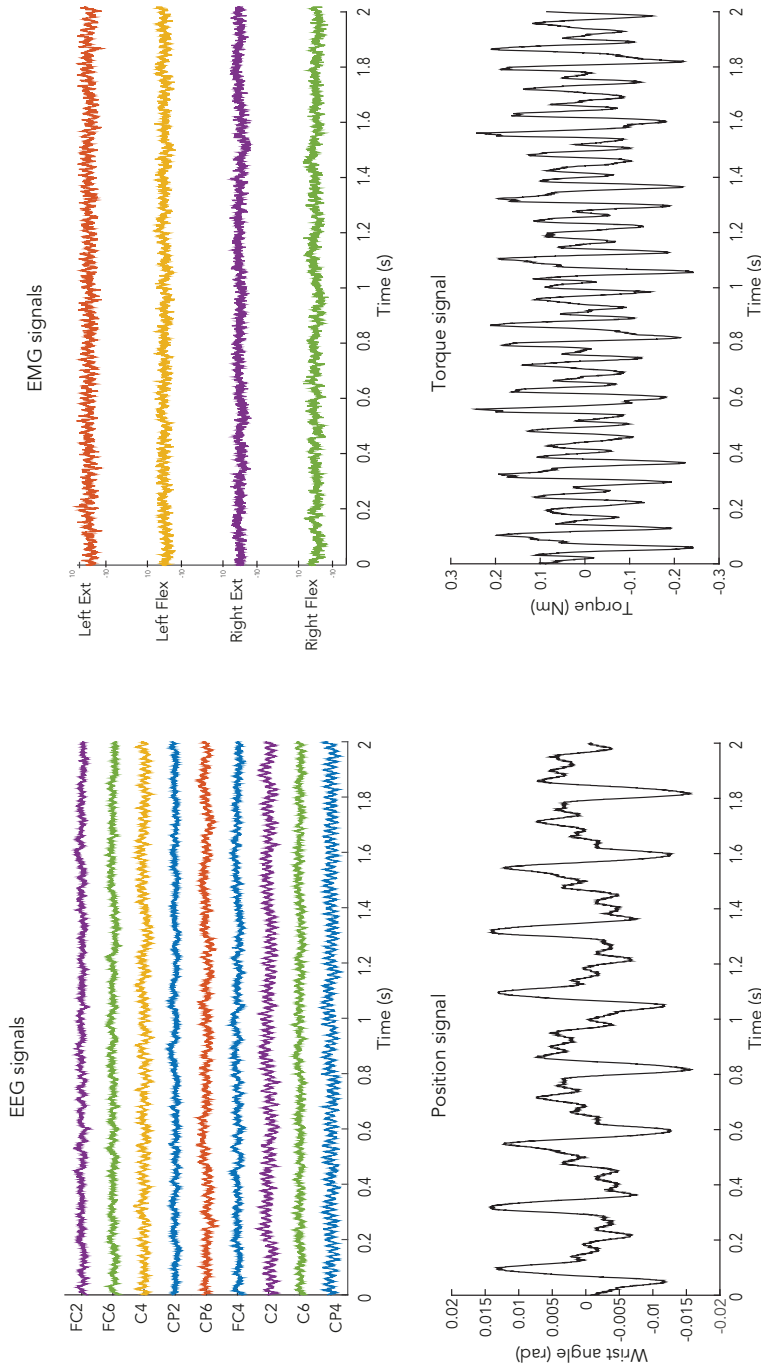
## Funding

This study was financially supported by a fellowship granted to C.G.M. Meskers (F2011(1)-25) from the Dutch brain foundation (Hersenstichting), the Netherlands, and a European advanced grant (ERC-AG-ID1, FP7-IDEAS-ERC, 291339).

## REFERENCES

- 1 Langhorne P, Bernhardt J, Kwakkel G. Stroke rehabilitation. *Lancet* 2011; **377**: 1693–702.
- 2 Kwakkel G, Kollen B, Twisk J. Impact of time on improvement of outcome after stroke. *Stroke* 2006; **37**: 2348–2353.
- 3 Winters C, Kwakkel G, Nijland R, van Wegen E. When does return of voluntary finger extension occur post-stroke? A prospective cohort study. *PLoS One* 2016; **11**: e0160528.
- 4 Winters C, van Wegen EEH, Daffertshofer A, Kwakkel G. Generalizability of the proportional recovery model for the upper extremity after an ischemic stroke. *Neurorehabil Neural Repair* 2015; **29**: 614–622.
- 5 Rothwell JC, Traub MM, Marsden CD. Influence of voluntary intent on the human long-latency stretch reflex. *Nature* 1980; **286**: 496–498.
- 6 Perenboom MJL, Van de Ruit M, De Groot JH, Schouten AC, Meskers CGM. Evidence for sustained cortical involvement in peripheral stretch reflex during the full long latency reflex period. *Neurosci Lett* 2015; **584**: 214–218.
- 7 Carey LM, Matyas TA, Oke LE. Sensory loss in stroke patients: effective training of tactile and proprioceptive discrimination. *Arch Phys Med Rehabil* 1993; **74**: 602–611.
- 8 Stolk-Hornsveld F, Crow JL, Hendriks EP, Van Der Baan R, Harmeling-van Der Wel BC. The Erasmus MC modifications to the (revised) Nottingham Sensory Assessment: a reliable somatosensory assessment measure for patients with intracranial disorders. *Clin Rehabil* 2006; **20**: 160–172.
- 9 Sullivan JE, Hedman LD. Sensory dysfunction following stroke: incidence, significance, examination, and intervention. *Top Stroke Rehabil* 2008; **15**: 200–217.
- 10 Connell LA, Tyson SF. Measures of sensation in neurological conditions: a systematic review. *Clin Rehabil* 2012; **26**: 68–80.
- 11 Kalogianni K, Daffertshofer A, van der Helm FCT, Schouten AC, de Munck JC, Kwakkel G et al. Disentangling somatosensory evoked potentials of the fingers: limitations and clinical potential. *Brain Topogr* 2018; **31**: 498–512.
- 12 Parkkonen E, Laaksonen K, Piitulainen H, Pekkola J, Parkkonen L, Tatlisumak T et al. Strength of ~20-Hz rebound and motor recovery after stroke. *Neurorehabil Neural Repair* 2017; **31**: 475–486.
- 13 Campfens SF, Schouten AC, van Putten MJ, van der Kooij H. Quantifying connectivity via efferent and afferent pathways in motor control using coherence measures and joint position perturbations. *Exp Brain Res* 2013; **228**: 141–153.
- 14 Campfens SF, Zandvliet SB, Meskers CGM, Schouten AC, van Putten MJAM, van der Kooij H. Poor motor function is associated with reduced sensory processing after stroke. *Exp Brain Res* 2015; **233**: 1339–1349.
- 15 Vlaar MP, Solis-Escalante T, Dewald JPA, van Wegen EEH, Schouten AC, Kwakkel G et al. Quantification of task-dependent cortical activation evoked by robotic continuous wrist joint manipulation in chronic hemiparetic stroke. *J Neuroeng Rehabil* 2017; **14**: 30.
- 16 Bernhardt J, Borschmann K, Boyd L, Carmichael ST, Corbett D, Cramer SC et al. Moving Rehabilitation Research Forward: Developing Consensus Statements for Rehabilitation and Recovery Research. *Neurorehabil Neural Repair* 2017; **31**: 694–698.
- 17 Stinear CM. Prediction of motor recovery after stroke: advances in biomarkers. *Lancet Neurol* 2017; **16**: 826–836.
- 18 Boyd LA, Hayward KS, Ward NS, Stinear CM, Rosso C, Fisher RJ et al. Biomarkers of stroke recovery: consensus-based core recommendations from the stroke recovery and rehabilitation roundtable. *Int J Stroke* 2017; **12**: 480–493.
- 19 Oostenveld R, Fries P, Maris E, Schoffelen J-M. FieldTrip: Open source software for advanced analysis of MEG, EEG, and invasive electrophysiological data. *Comput Intell Neurosci* 2011; **2011**: e156869.
- 20 Bortel R, Sovka P. Approximation of statistical distribution of magnitude squared coherence estimated with segment overlapping. *Signal Processing* 2007; **87**: 1100–1117.
- 21 Duncan PW, Propst M, Nelson SG. Reliability of the Fugl-Meyer assessment of sensorimotor recovery following cerebrovascular accident. *Phys Ther* 1983; **63**: 1606–1610.

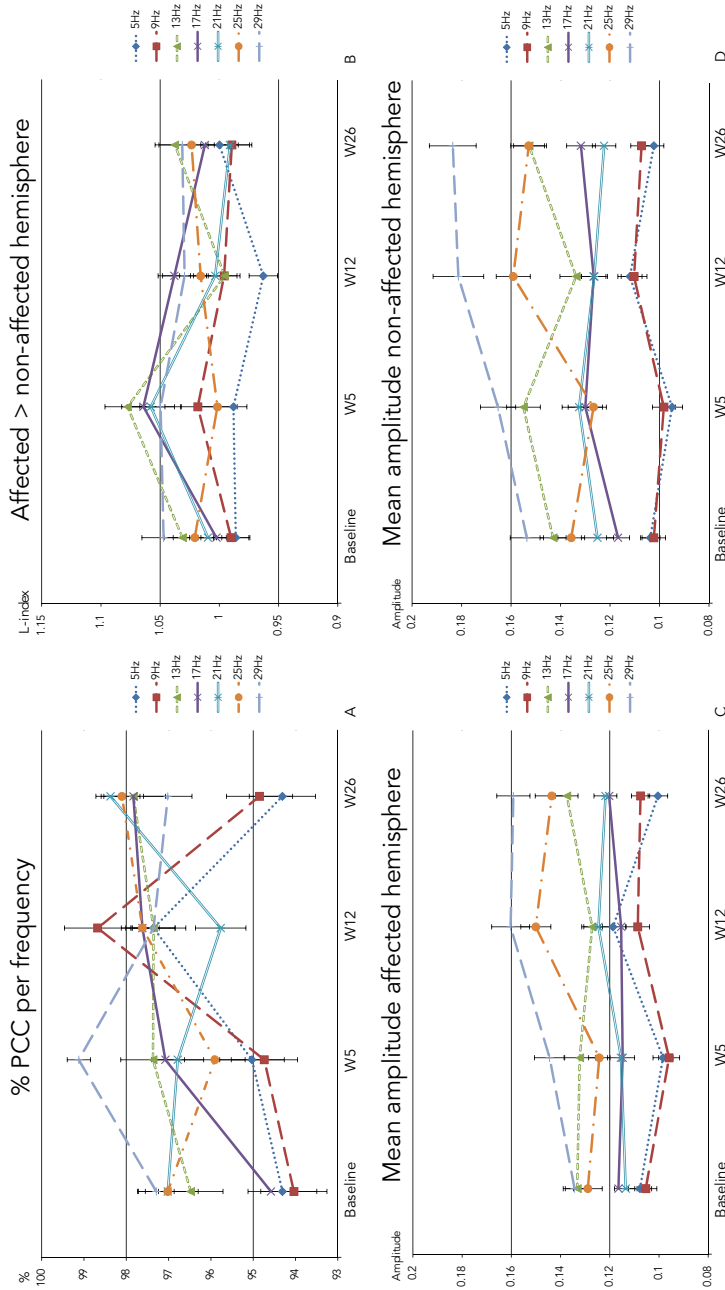
- 22 Prabhakaran S, Zarahn E, Riley C, Speizer A, Chong JY, Lazar RM et al. Inter-individual variability in the capacity for motor recovery after ischemic stroke. *Neurorehabil Neural Repair* 2008; **22**: 64–71.
- 23 Budtz-Jorgensen E, Keiding N, Grandjean P, Weihe P. Confounder selection in environmental epidemiology: assessment of health effects of prenatal mercury exposure. *Ann Epidemiol* 2007; **17**: 27–35.
- 24 van Kordelaar J, van Wegen E, Kwakkel G. Impact of time on quality of motor control of the paretic upper limb after stroke. *Arch Phys Med Rehabil* 2014; **95**: 338–344.
- 25 Buma FE, Lindeman E, Ramsey NF, Kwakkel G. Review: functional neuroimaging Studies of early upper limb recovery after stroke: a systematic review of the literature. *Neurorehabil Neural Repair* 2010; **24**: 589–608.
- 26 Laaksonen K, Kirveskari E, Makela JP, Kaste M, Mustanoja S, Nummenmaa L et al. Effect of afferent input on motor cortex excitability during stroke recovery. *Clin Neurophysiol* 2012; **123**: 2429–2436.
- 27 Nicolo P, Rizk S, Magnin C, Pietro M Di, Schnider A, Guggisberg AG. Coherent neural oscillations predict future motor and language improvement after stroke. *Brain* 2015; **138**: 3048–3060.
- 28 Ward NS. Restoring brain function after stroke - bridging the gap between animals and humans. *Nat Rev Neurol* 2017; **13**: 244–255.
- 29 Kwakkel G, Lannin NA, Borschmann K, English C, Ali M, Churilov L et al. Standardized measurement of sensorimotor recovery in stroke trials: Consensus-based core recommendations from the Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2017; **12**: 451–461.
- 30 Vlaar MP, Birpoutsoukis G, Lataire J, Schoukens M, Schouten AC, Schoukens J et al. Modeling the Nonlinear Cortical Response in EEG Evoked by Wrist Joint Manipulation. *IEEE Trans Neural Syst Rehabil Eng* 2018; **26**: 205–215.
- 31 Yang Y, Solis-Escalante T, van der Helm FCT, Schouten AC. A generalized coherence framework for detecting and characterizing nonlinear interactions in the nervous system. *IEEE Trans Biomed Eng* 2016; **63**: 2629–2637.
- 32 Campfens SF, Meskers CGM, Schouten AC, van Putten MJAM, van der Kooij H. Stretch Evoked Potentials in Healthy Subjects and After Stroke: A Potential Measure for Proprioceptive Sensorimotor Function. *IEEE Trans Neural Syst Rehabil Eng* 2015; **23**: 643–654.
- 33 Hope TMH, Friston K, Price CJ, Leff AP, Rotshtein P, Bowman H. Recovery after stroke: not so proportional after all? *Brain* 2019; **142**: 15–22.



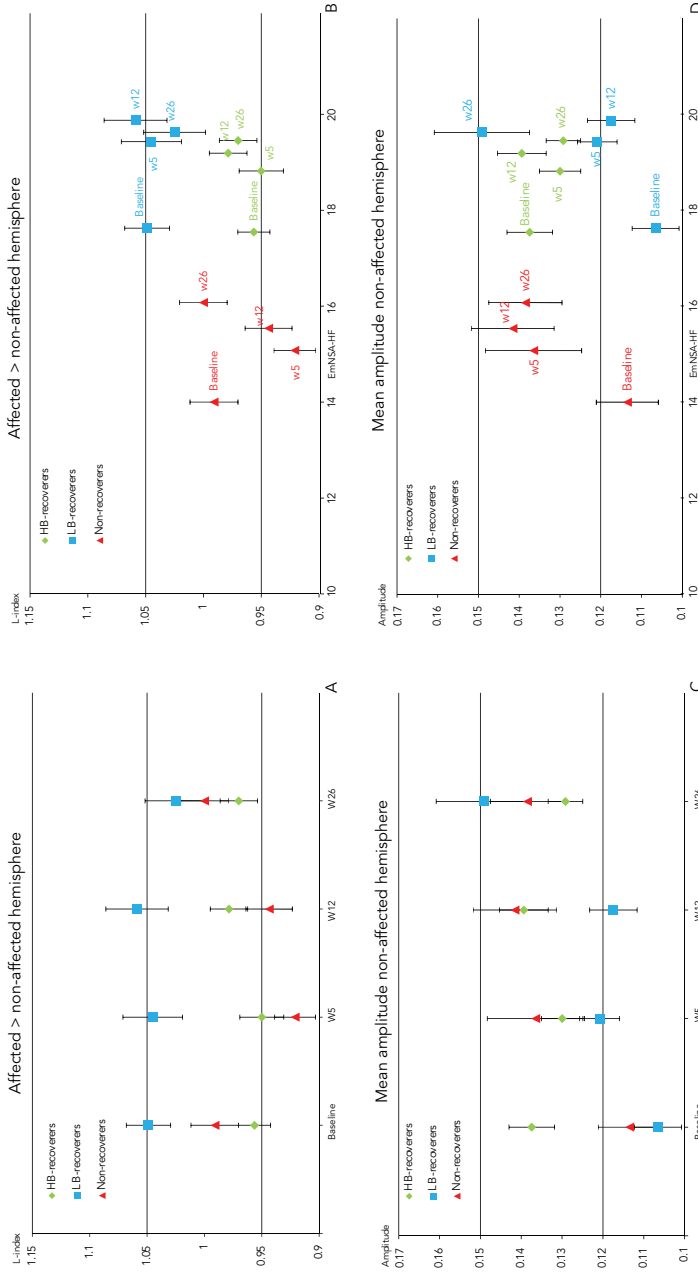
**Supplementary Figure S5.1** | Display of 2 seconds representative data.

Signals were high pass filtered with a second order butterworth filter with a cutoff of 1Hz to remove the linear trend. The EEG and EMG signals are displayed on a  $-20\mu\text{V}$  to  $20\mu\text{V}$  range. Panel A: EEG signals of the nine channels overlapping the motor cortex area from which position-cortical coherence was calculated. The patient had a right hemisphere lesion. Panel B: EMG signals of the right and left m. flexor carpi radialis and the m. extensor carpi radialis longus. The left hand of the patient was concurrently perturbed. Panel C: 2 seconds of the measured torque signal of the left hand during the imposed position perturbation. Panel D: 2 seconds of the measured torque signal when the position perturbation signal was imposed onto the left hand of the patient.



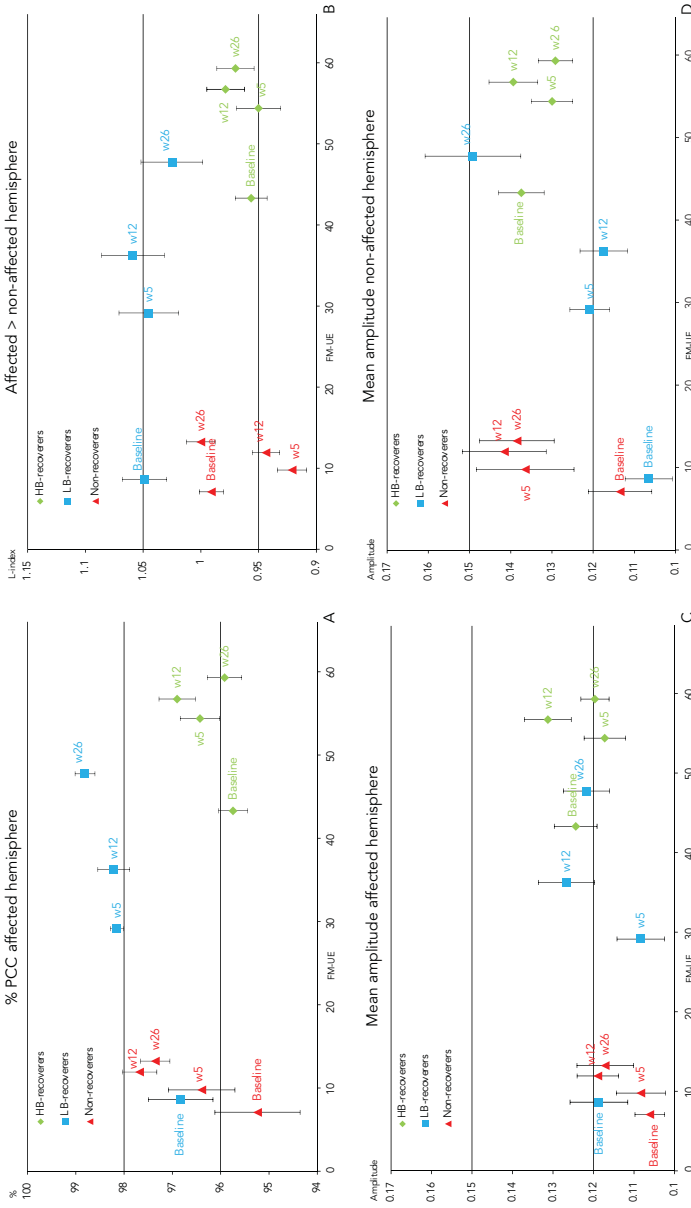


**Supplementary Figure S5.2** | PCC parameters over time per perturbed frequency. position-cortical coherence (PCC) calculated for the electrodes overlapping the sensorimotor cortex displayed per perturbed frequency at baseline and at 5, 12 and 26 weeks post-stroke. The perturbation was applied to the affected arm. Error bars indicate the standard error of the mean. Panel A: Percentage of significant PCC per frequency of the electrodes over the affected hemisphere. Panel B: The lateralisation shows if the PCC was more lateralised towards the affected ( $L > 1$ ) or towards the non-affected ( $L < 1$ ) sensorimotor cortex per frequency. Panel C: Mean amplitude per frequency of PCC of all electrodes overlapping the sensorimotor cortex of the affected hemisphere. Panel D: Mean amplitude per frequency of PCC of all electrodes overlapping the sensorimotor cortex of the non-affected hemisphere.



**Supplementary Figure S5.3** | Amplitude of PCC in the non-affected hemisphere and the lateralization index in the recovery groups, over time and in relation with EmNSA-HF. position-cortical coherence (PCC) calculated for the electrodes overlapping the sensorimotor cortex for high-baseline recoverers (HB-recovers), low-baseline recoverers (LB-recovers) and non-recovers, at baseline and at 5, 12 and 26 weeks post-stroke. The Erasmus modification of the Nottingham sensory assessment of the hand and fingers (EmNSA-HF) was measured at the same time points. Error bars indicate the standard error of the mean. Panel A: The lateralisation shows if the PCC was more lateralised towards the affected ( $L > 1$ ) or towards the non-affected ( $L < 1$ ) sensorimotor cortex. Panel B: L-index versus the EmNSA-HF. Panel C: Mean amplitude of PCC of all electrodes overlapping the sensorimotor cortex of the unaffected hemisphere. Panel D: Mean amplitude of PCC of the unaffected hemisphere versus the EmNSA-HF.





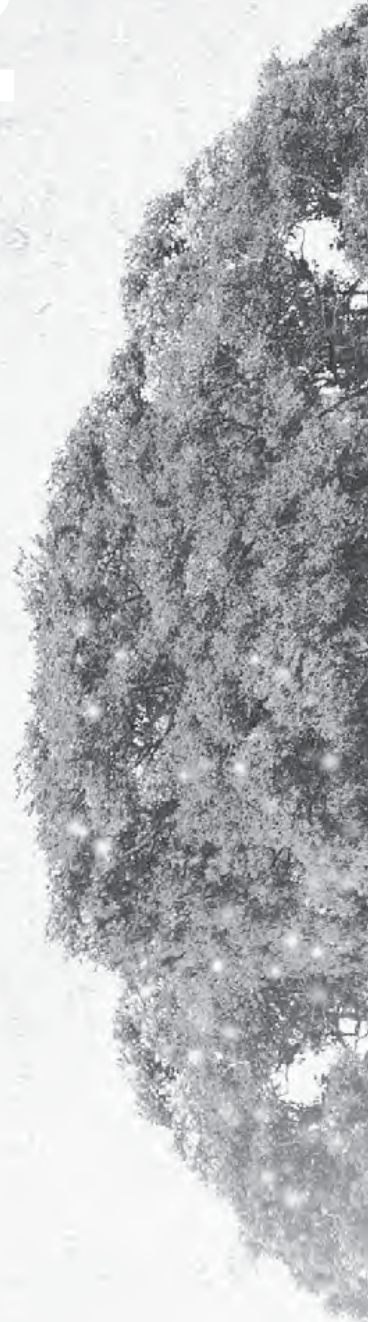
**Supplementary Figure S5.4** | Relation between the FM-UE and the PCC parameters for the different recovery groups.

Relation between the Fugl-Meyer motor assessment of the upper extremity (FM-UE) and the four position-cortical coherence (PCC) measures. PCC was calculated for the electrodes overlapping the sensorimotor cortex for high-baseline recoverers (HB-recovers), low-baseline recoverers (LB-recovers) and non-recovers, at baseline and at 5, 12 and 26 weeks post-stroke. The perturbation was applied to the affected arm. Error bars indicate the standard error of the mean. Panel A: Percentage of significant PCC of the electrodes over the affected hemisphere. Panel B: The lateralisation shows if the PCC was more lateralised towards the affected ( $L > 1$ ) or towards the non-affected ( $L < 1$ ) sensorimotor cortex. Panel C: Mean amplitude of PCC of all electrodes overlapping the sensorimotor cortex of the affected hemisphere. Panel D: Mean amplitude of PCC of all electrodes overlapping the sensorimotor cortex of the non-affected hemisphere.



# PART 2

Modulation of sensorimotor  
recovery after stroke











# Chapter 6

Short term effects  
of cerebellar tDCS  
on standing balance  
performance in patients  
with chronic stroke and  
healthy age matched  
elderly

Sarah B. Zandvliet  
Carel G.M. Meskers  
Gert Kwakkel  
Erwin E.H. van Wegen

## ABSTRACT

**Introduction:** Transcranial direct current stimulation (tDCS) may serve as an adjunct approach in stroke rehabilitation. The cerebellum could be a target during standing balance training due to its role in motor adaptation. We tested whether cerebellar tDCS can lead to short term effects on standing balance performance in patients with chronic stroke.

**Methods:** 15 patients with a chronic stroke were stimulated with anodal stimulation on the: contra-lesional cerebellar hemisphere, ipsi-lesional cerebellar hemisphere or with sham stimulation, for 20 minutes with 1.5 mA in three sessions in randomized order. 10 healthy controls participated in two sessions with cerebellar stimulation ipsi-lateral to their dominant leg or with sham stimulation. During stimulation subjects performed a medio-lateral postural tracking task on a force platform. Standing balance performance was measured directly before and after each training session in several standing positions. Outcomes were centre of pressure (CoP) amplitude and its standard deviation, velocity and its standard deviation and range, subsequently combined into a CoP composite score (comp-score) as a qualitative outcome parameter.

**Results:** In the patient group, a decrease in comp-score in the tandem position was found after contra-lesional tDCS:  $\beta=-0.25$ ,  $CI=-0.48$  to  $-0.03$ ,  $P=0.03$ . No significant differences in demographics and clinical characteristics were found between patients that responded ( $N=10$ ) and patients that did not respond ( $N=5$ ) to the stimulation.

**Discussion:** Contra-lesional cerebellar tDCS shows promise for improving standing balance performance. Exploration of optimal timing, dose and the relation between qualitative parameters and clinical improvements, are needed to establish whether tDCS can augment standing balance performance after stroke.

---

## INTRODUCTION

Recovery of standing balance after stroke is a key factor in regaining independence in activities of daily living (ADL) and preventing fall-events <sup>1</sup>. A meta-analysis of interventions aimed to improve standing balance did not indicate superiority of a certain training method, suggesting the need for more effective interventions post-stroke <sup>2</sup>.

Recently, non-invasive transcranial direct current stimulation (tDCS) has emerged as an innovative, promising approach in stroke rehabilitation <sup>3</sup>. tDCS may prime the brain before or during a therapeutic intervention, providing potential to augment the positive learning effects of task specific training, the idea being that such combined peripheral and central input enhances synaptic plasticity and skill relearning <sup>4</sup>. Recent literature however, reports inconsistent findings on improvement in motor performance when measured with clinical scales in patients with chronic stroke, suggesting that, if any effect exists at all, tDCS interventions may only induce subtle changes <sup>5</sup>. In addition, clinical outcomes such as gait speed and the Berg balance scale, are not able to delineate between 'true neurobiological repair' and behavioural compensation strategies <sup>6</sup>. Therefore, kinematic and kinetic measures are recommended in stroke recovery trials to demonstrate possible effects of tDCS in terms of quality of motor performance <sup>7</sup>. One may also argue that the subtle effects of tDCS, which are believed to enhance Hebbian and non-Hebbian learning processes by mechanisms of long term potentiation (LTP) and long term depression (LTD)-like plasticity may benefit most in those brain areas that are responsible for learning-dependent motor control such as the cerebellum <sup>8</sup>.

The cerebellum is known to be involved in error based motor learning, also referred to as motor adaptation <sup>9</sup>. LTD-like plasticity of Purkinje cells is associated with learning this Hebbian process is mediated by simultaneous activation of parallel fibers and climbing fibers that give input to error signals in motor control to the cortex <sup>10-12</sup>. Balance performance can be seen as adaptation of posture <sup>13</sup>, and the cerebellar hemispheres play a specific role in motor adaptation <sup>14,15</sup>. A strong M1-cerebellar connection also results in more accurate movement endpoints, emphasizing the crucial role of the cerebellum in motor adaptation <sup>9,13,16</sup>. The more medial flocculonodular lobe of the cerebellum is directly linked to postural balance <sup>17</sup>, but can likely not be targeted with tDCS due to its anterior location, while the hemispheres can be targeted with tDCS <sup>18</sup>.

Cathodal stimulation of a cerebellar hemisphere leads to a decrease in cerebellar brain inhibition, likely via an enhancement of LTD of Purkinje cells <sup>19,20</sup>. In healthy subjects, cathodal stimulation, of the cerebellum has been found to lead to improvement of balance performance in the study of Inukai et al. <sup>21</sup>, while Foerster et al. <sup>22</sup> found an impairing

effect. Anodal stimulation has been found to lead to significant improvements in motor adaptation and balance performance in several studies in healthy subjects, while others found no added value of stimulation<sup>15,21-25</sup>. The potential benefits of cerebellar tDCS in stroke patients with balance impairments, has to date not been investigated.

It has been suggested that anodal cerebellar tDCS (cb-tDCS) enlarges the population of activated Purkinje cells, leading to a larger involvement of the cerebellum in the executed motor task<sup>24</sup>. More recent animal studies also showed that synaptic based forms of learning cannot take place in the absence of LTP of Purkinje cells, suggesting a complex interplay of LTD and LTP in the cerebellum<sup>8,26-28</sup>. Bearing in mind the function of the cerebellum, as controller of temporal and spatial accuracy, it can be argued that both Hebbian and anti-Hebbian processes play a role in the cerebellum and optimisation of these processes can lead to an enhancement of motor adaptation. It is plausible that a positive effect of stimulation can therefore only be found if there is a need for improvement of balance performance, which is unlikely in healthy young adults, however there is a substantial clinical problem in patients with a stroke<sup>1,2,25</sup>.

In this proof of concept study we investigated for the first time the short term effects of anodal cb-tDCS applied on both the ipsi-lesional as well as the contra-lesional cerebellar hemisphere as compared to sham stimulation during a postural training task in patients with chronic stroke and healthy age-matched controls. Since clinical tests are not sensitive enough to record the qualitative aspects of balance performance and with that, unable to record the subtle changes that can be expected from a single session of tDCS, the effects are measured with kinetic parameters. We hypothesised that both anodal stimulation conditions normalize standing balance performance measured with centre of pressure (CoP) derived parameters in patients with stroke. Normalisation of standing balance performance was defined as a decrease in CoP parameters in patients with a stroke. The largest effect on standing balance performance was expected in the most difficult task, a (semi-)tandem stance, due to the expected room for improvement via motor adaptation, especially in the group of patients with a stroke.

## METHODS

Measurements took place at the rehabilitation department of the VU university medical center. All procedures performed in this study involving human participants were in accordance with the ethical standards of the institutional research committee and with the 1964 Helsinki declaration and its later amendments and approved under reference: NL52021.029.15. All subjects gave their written informed consent.

## Subjects

Fifteen patients with chronic stroke (>6 months post-stroke) and ten age-gender matched healthy controls were enrolled in this study. Patients had to meet the following criteria to participate:

(1) A first ever ischemic or haemorrhagic lesion excluding lesions of the cerebellum as verified by CT or MRI scan; (2) decreased standing balance performance as determined by a score of <56 points on the Berg balance scale (BBS).

In addition, all patients and healthy controls had to meet the following additional criteria:

(1) age  $\geq 18$ ; (2) normal or corrected to normal vision with an optical aid; (3) able and sufficiently motivated to perform the required tests and interventions; (4) no metallic implants near to the side of stimulation; (5) no orthopaedic limitations that interfere with the study; (6) no cranial bone defects; (7) no history of epileptic seizures; (8) no severe psychiatric disorder (e.g. bipolar or psychotic disorder or suicidality); (9) no signs of depression (hospital anxiety and depression scale, HADS, sub score D <7)<sup>29</sup>; (10) sufficient cognitive function (mini mental state examination, MMSE  $\geq 19$ ); (11) no sensory impairments (prior to the ischemic lesion, in case of patients); (12) no diagnosed diseases of the vestibular system; (13) absence of additional therapy focussing on standing balance improvement during the time period in which the measurements took place; (14) no history of disease, condition, event or use of medication that interfered with the study.

## Protocol

All subjects first participated in an intake and clinical assessment session. After which they returned for respectively 2 (healthy controls) or 3 (patients) sessions, in which they had to perform a medio-lateral postural tracking task while being stimulated with anodal cb-tDCS. Directly before and after this tracking task with cb-tDCS, standing balance performance was assessed during several quiet standing tests. These sessions were minimally 1 and maximally 2 weeks apart. For a schematic overview of the protocol, see Figure 6.1.

## Tracking task

Two dots were presented on a video screen located at eye height for the tracking task (Figure 6.2, panel C). One dot represented a moving target, which the subject was asked to follow as precisely as possible with the second tracking dot, representing their CoP measured with a force plate. The CoP signal was low pass filtered with a second order Butterworth filter at 10 Hz (D-flow, Motek, Amsterdam, the Netherlands). Four repetitions

of the tracking task of 3 minutes each were performed by each subject. In two of those repetitions, the target moved in predictable manner with an increasing velocity in eight blocks of 20 seconds from 0.16 to 1.28 cm/sec. During the two other repetitions, the target moved in a pseudo-random manner with 16 blocks of 10 seconds with a velocity between 0.16 and 1.28 cm/sec. Those repetitions were therefore considered to be unpredictable in terms of velocity. Subjects received feedback on their performance after each repetition. Performance was defined as the percentage of the time of accurate overlap of the target and tracking dot during the eight different velocities. Subsequently, subjects could rest as long as needed between repetitions.

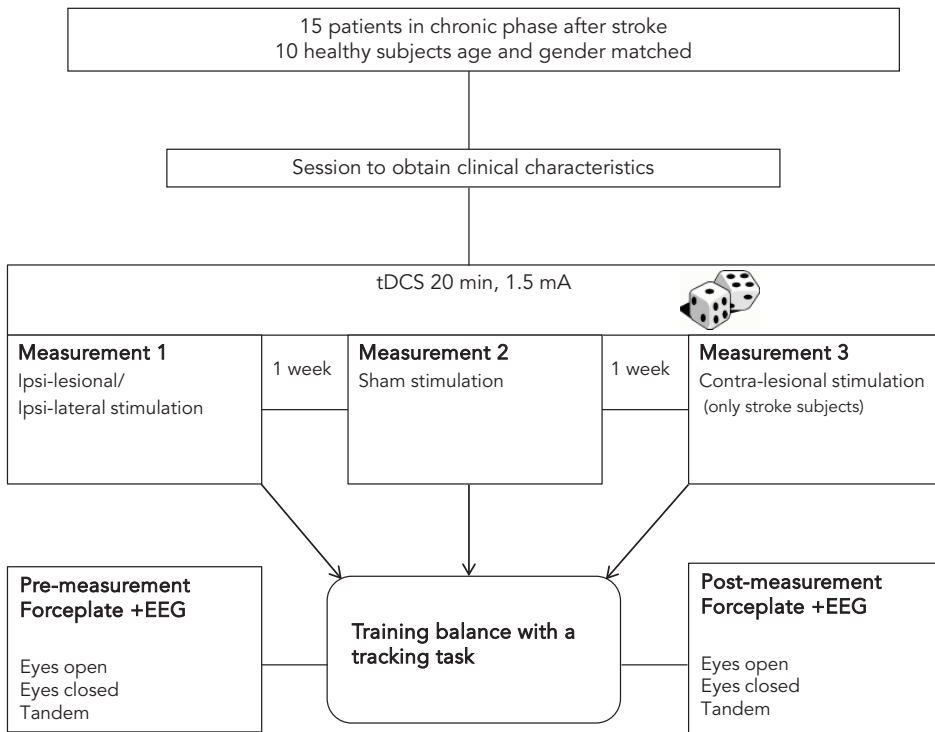


Figure 6.1 | Overview of the experimental protocol.

Patients were stimulated in one session at the contra-lesional side, and in one session at the ipsi-lesional side. Sham stimulation was applied to the contra-lesional side. Healthy controls only had two sessions both ipsi-lateral to their dominant leg with either real or sham stimulation.

## Performance on the tracking task

To evaluate a possible learning effect of the tracking task, performances of the last two repetitions were averaged over all frequencies. This included one repetition with a predictable and one with an unpredictable moving target.

## tDCS

During the tracking task cb-tDCS was delivered by a battery-driven portable stimulator (Starstim®, Neuroelectronics, Barcelona, Spain) through 3.14 cm<sup>2</sup> electrodes filled with conducting gel, with a direct 1.5 mA current. The anodal electrode was placed 3 centimetres lateral of the inion. Two cathodal electrodes were placed on the ipsi-lateral buccinators muscles (Figure 6.2, panel A). Anodal stimulation was applied for 20 minutes with 1.5 mA in all sessions. During the first 3 minutes of stimulation, subjects were sitting on a chair to get acquainted with the stimulation while the tracking task was explained. For patients, three experimental sessions, with minimally 1 week and maximally 2 weeks apart were performed: 1) anodal cb-tDCS on contra-lesional side, 2) anodal cb-tDCS on the ipsi-lesional side and 3) Sham cb-tDCS on the contra-lesional side. In Sham tDCS the current was automatically switched off after 30 seconds and switched on again for the last 30 seconds of the 20 minutes stimulation period. Healthy controls only had two sessions, both ipsi-lateral to their dominant leg, with either anodal or sham stimulation. The order of the sessions was randomized. The stimulation was always finished before subjects finished their fourth repetition of the tracking task. The dominant leg was determined by the preferred leg used and better performance on the lateralized items of the BBS (i.e. looking over the shoulder, turning, tandem stance and one leg stance).

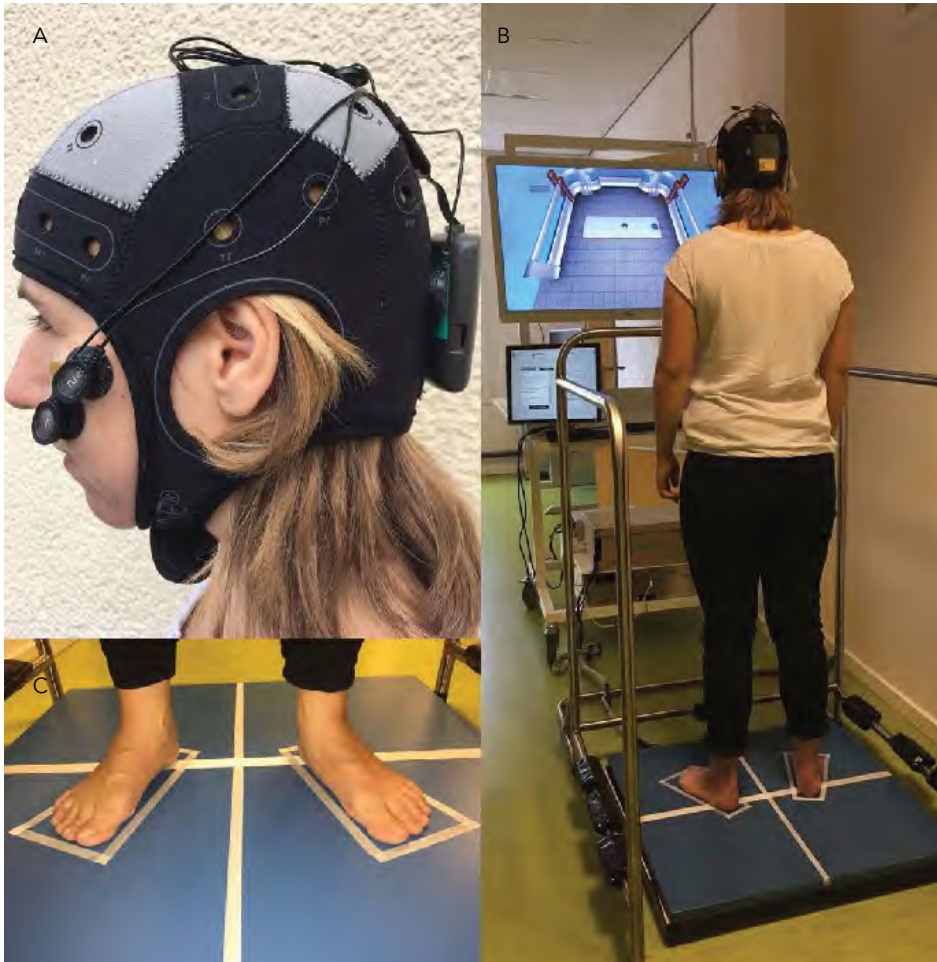
## Standing balance performance assessment

Before and after the stimulation, standing balance performance was assessed during several quiet standing positions. Ground reaction forces were measured during these positions with a sample frequency of 1000 Hz using an 80x80 cm force plate (Motek, Amsterdam, the Netherlands). The analogue signals were converted with a 16bit analogue-to-digital converter with a 10 Volt range (National Instruments, Austin, USA).

Subjects were asked to stand quietly in three sequential standing positions; 1) with eyes open, 2) eyes closed and 3) in the most challenging, subject specific, (semi-)tandem stance, on a force plate during 5 repetitions of 1 minute. Subjects rested for a minimum of 30 seconds in between standing tests. The rest period was extended upon the subjects' request to avoid fatigue. In standing position 1 and 2, subjects stood barefoot with their



arms relaxed, alongside the trunk if possible, with their feet at hip width in 9 degrees exorotation (Figure 6.2, panel B). During the tandem stance, subjects were asked to stand in the most difficult position which they could hold for one minute as determined in the first clinical assessment session. The foot positions of the first session were marked and measured to keep foot placement the same in the consecutive sessions.



**Figure 6.2 |** Overview of experimental setup.

Panel A: Cerebellar transcranial direct current stimulation was delivered by a portable stimulator with 1.5 mA current for 20 minutes in all sessions.

Panel B: Subject performing the tracking task. The blue dot represented a moving target, which the subject was asked to follow as precisely as possible with the grey tracking dot, representing their centre of pressure measured with the force plate.

Panel C: Subjects stood with their feet at hip width in 9 degrees exorotation for 5 times 1 minute per standing position. The position was marked for repositioning after rest.

At four time points during a session, subjects were asked to indicate their level of headache, nausea, fatigue and depressed mood on a visual analogue scale (VAS) from 0–100 mm, in order to track commonly reported side effects of tDCS <sup>30</sup>.

All subjects were told at the end of all measurements that during one of the sessions a placebo stimulation was given and asked if they could indicate which session it was.

### Clinical assessments

The clinical measures were performed by the researcher according to recommended guidelines 2 before the sessions with tDCS. The following assessments were performed: Berg balance scale (BBS) <sup>31</sup>, functional reach task (FRT) <sup>32</sup>, timed up and go (TUG) <sup>33</sup>, fall efficacy scale (FES) <sup>34</sup>, fall history, Erasmus modification of the Nottingham sensory assessment of the lower extremity (EmNSA-LE) <sup>35</sup>. For healthy controls the dominant leg was determined by the preferred leg used and better performance on the lateralized items of the BBS (i.e. looking over the shoulder, turning, tandem stance and one leg stance). The most difficult position for subjects to hold for more than 30 seconds was determined with an ordinal scale ranging from: 1) full tandem stance with non-paretic leg/dominant in front, 2) non-paretic/dominant leg a step ahead, 3) non-paretic/dominant leg half a step ahead, 5) full tandem stance with paretic leg/non-dominant in front, 6) paretic/non-dominant leg a step ahead, 7) paretic/non-dominant leg half a step ahead or 8) feet placed together.

Stroke patients were also assessed with the: Fugl-Meyer motor assessment of the lower extremity (FM-LE) <sup>36</sup>, motricity index (MI) <sup>37</sup>, Nottingham extended activities of daily living index (NEADL) <sup>38</sup>, and O-letter cancellation test (LCT) <sup>39</sup>.

### Data analysis and pre-processing

Recorded data were processed using MATLAB 2012a (the MathWorks, Inc., Natick, MA, USA). Statistical analysis was performed using IBM SPSS statistics version 22 (IBM corporation, Armonk, NY, USA).

### Outcome parameters

Force plate data was low pass filtered with a second order Butterworth filter with a cutoff frequency of 10 Hz, after which the CoP in both the anterior-posterior (AP) and the medio-lateral (ML) direction was calculated. The middle 50 seconds of each trial were used for further analysis. The signals were linear detrended with a period of 20 seconds. The following parameters were computed from the sum of the vectors of the CoP in the AP and

ML direction: the mean amplitude of the CoP (ACoP) calculated as the root mean squared distance from the mean CoP, its standard deviation represents the amplitude's variability (varCoP), the velocity of the CoP (VCoP) calculated by the sum of the distance between sequential points divided by its length, its standard deviation represents the velocity's variability (varVCoP) and the range determined as the maximal difference between any two points of the time series. A CoP composite score of the above mentioned parameters was calculated as a comprehensive outcome parameter representing qualitative aspects of standing balance performance<sup>40</sup>. Each parameter, calculated for ML and AP direction separately, was transformed to a z-score calculated over all parameters, separately for the stroke patients and the healthy controls. This transformation leads to a mean of 0 with a standard deviation of 1, making it possible to average the ten transformed parameters per standing position into a CoP composite-score (comp-score).

### **Statistical analysis**

Normality was checked by visual inspection of the probability distribution (q-q plot) and the box plot. A Shapiro-Wilks test was performed on the data or the residual when appropriate. When the assumptions of normality were not met, a natural log transformation was applied after which normality was checked again. In case this transformation was not sufficient, or in case of ordinal or nominal data, the non-parametric equivalent of the below mentioned tests was used. The significance level  $\alpha$  was set two-tailed at 0.05.

CoP baseline differences between healthy controls and patients were analysed with an independent t-test with a Bonferroni correction for multiple testing per standing position. To correct for differences in sample size, Hedges'g (g) was used to calculate effect sizes, when significant differences were found.

A generalized estimating equation (GEE) model, with a correction for baseline CoP comp-score, was used to establish an association between the stimulation conditions on CoP comp-scores for patients and healthy control separately. The model was also tested for confounding of randomization order. If the  $\beta$ -values for the stimulation conditions changed with more than 10%, this was considered an improvement of the model. In case of a significant association between post-measurement CoP comp-score and cb-tDCS, the five parameters from which the CoP comp-score was constructed were evaluated separately.

### ***Performance on the tracking task***

A GEE model was used to evaluate the improvement of performance on the tracking task over time. Stimulation condition was added as a possible confounder to examine if performance was influenced by cb-tDCS.

## Responders and non-responders

To investigate the differences in response to cb-tDCS between subjects (interindividual response), a distinction was made between patients who showed a response on the CoP comp-score and patients that did not. A subject was defined as a 'responder' when a change in CoP comp-score (post – pre) for the tandem stance was more than one standard deviation larger in the contra-lesional stimulation or the ipsi-lesional condition compared to the sham condition. Differences in baseline CoP comp-score, fatigue and clinical and demographic characteristics between responders and non-responders were analysed with independent t-tests.

## RESULTS

### Procedures

All subjects completed the experimental protocol. One patient was not able to perform 5 repetitions of each static assessment of standing balance performance due to fatigue. Three repetitions of each assessment were performed instead.

Two patients were able to perform a semi-tandem stance for 60 seconds with the non-paretic leg ahead of the paretic leg, ten patients were able to perform a semi-tandem stance with the non-paretic leg a small step in front of the paretic leg, and 3 patients were able to perform a semi-tandem stance with the paretic leg a small step in front of the non-paretic leg. Nine healthy controls were able to perform a tandem stance with the dominant leg ahead, one healthy control performed a semi-tandem stance with the dominant leg ahead.

### Successfulness of blinding procedures

When asked to indicate the sham condition at the end of the protocol, 4 out of 10 healthy controls answered correctly, 5 incorrectly and 1 could not make a choice. From the patients 7 answered correctly, 6 incorrectly and 2 patients could not make a choice.

### Possible side effects, fatigue

No subjects reported headaches or nausea during any of the sessions. Three healthy controls reported to be somewhat fatigued but no differences in VAS on fatigue between sham: median=0, inter quartile ranges (IQR)=0–1.5 and cb-tDCS median=0, IQR=0–0, were found,  $z=-0.54$ ,  $P=0.60$ . 11 patients reported a higher VAS on fatigue after stimulation

but no differences were found between the sham median=11, IQR=0–30. contra-lesional: median=10, IQR=0–26 and ipsi-lesional median=13, IQR=0–21,  $\chi^2=0.98$ ,  $P=0.61$  conditions. Only one patient reported 70 out of 100 points on the VAS for depressed mood at the start of one of the sessions, no increase was reported after stimulation.

### Baseline differences between patients and healthy controls

Characteristics of both groups are displayed in Table 6.1. Healthy subjects and patients did not differ significantly in age, weight and height. Patients had a lower score on the BBS median=50, IQR=48–53, as well as on the EmNSA-LE: median=38 IQR=34–39 and were slower on the TUG: mean=14.3, SD=7.9, compared to healthy controls, BBS median=56, IQR=56–56,  $P<0.01$ , EmNSA-LE median=39, IQR=39–40,  $P=0.02$ , TUG mean=6.1, SD=0.99,  $P<0.01$ .

The baseline CoP parameters are displayed in Figure 6.3. Patients showed significant larger excursion on all baseline CoP parameters for the eyes open; ACoP mean healthy=2.90 mm, SD=0.47, mean patients=4.17 mm, SD=1.53, CI of the mean difference =-2.16–0.38,  $t(17.6)=-3.01$ ,  $g=1.00$ , varCoP: healthy=3.67 mm, SD=0.58, patients=5.35 mm, SD=1.99, CI=-2.83–0.53,  $t(17.3)=-3.07$ ,  $g=1.02$ , range: healthy=20.68 mm, SD=0.22, patients=32.21 mm, SD=0.42, CI=-18.88–4.17,  $t(16.4)=-3.31$ ,  $g=31.37$ , VCoP: median healthy=7.10 mm/s, IQR=5.88–7.81, median patients=10.78 mm/s, IQR=8.80–15.48, CI=0.43–0.78, ratio due to log transformation,  $t(23)=0.02$ ,  $g=1.71$ , varVCoP: median healthy= 5.79 mm/s, IQR=5.79–6.62, patients=8.81 mm/s, IQR=7.76–13.87, CI=0.40–0.75, ratio due to log transformation,  $t(23)=0.02$ ,  $g=1.57$ , all  $P<0.05$ .

This was also the case for the eyes closed position when compared to healthy-age-matched controls; ACoP: mean healthy=3.66 mm, SD=0.65, mean patients=5.71 mm, SD=2.02, CI=-3.23–0.88,  $t(18)=-3.67$ ,  $g=1.22$ , varCoP: healthy=4.66 mm, SD=0.83, patients=7.21 mm, SD=2.57, CI=-4.06–1.07,  $t(18)=-3.60$ ,  $g=1.20$ , range: healthy=26.49 mm, SD=5.23, patients=42.16 mm, SD=15.20, CI=-24.59–6.74,  $t(18.5)=-3.68$ ,  $g=1.23$ , VCoP: healthy=11.62 mm/s, SD=2.28, patients=20.44 mm/s, SD=8.91, CI=-13.91–3.73,  $t(16.6)=-3.66$ ,  $g=1.20$ , varVCoP: healthy=9.42 mm/s, SD=1.94, patients=17.26 mm/s, SD=7.56, CI=-12.16–3.51,  $t(16.6)=-3.83$ ,  $g=1.26$ , all  $P<0.05$ . For the (semi-)tandem stance position no significant differences were found between patients and healthy controls; ACoP: median healthy=5.17 mm, IQR=4.65–6.34, median patients=5.86 mm, IQR=4.65–7.89, ratio due to log transformation, CI=0.63–1.15,  $t(23)=0.33$ , varCoP: median healthy=6.55 mm, IQR=5.80–8.45, median patients=7.31 mm, IQR=5.82–10.09, ratio due to log transformation, CI=0.63–1.15,  $t(23)=0.33$ , range: median healthy=40.29

mm IQR=36.65–55.94, median patients=47.72 mm, IQR=33.14–64.73, ratio due to log transformation, CI=0.64–1.17,  $t(23)=0.37$ , median VCoP: healthy 30.25=mm/s, IQR=26.22–32.48, median patients=26.33 mm/s, IQR=23.90–36.83,  $U=61$ ,  $z=-0.78$ , varVCoP: median healthy 26.24=mm/s, IQR=21.40–28.96, median patients=23.84 mm/s IQR=19.47–31.16,  $U=71$ ,  $z=-0.22$ , all,  $P>0.5$ .

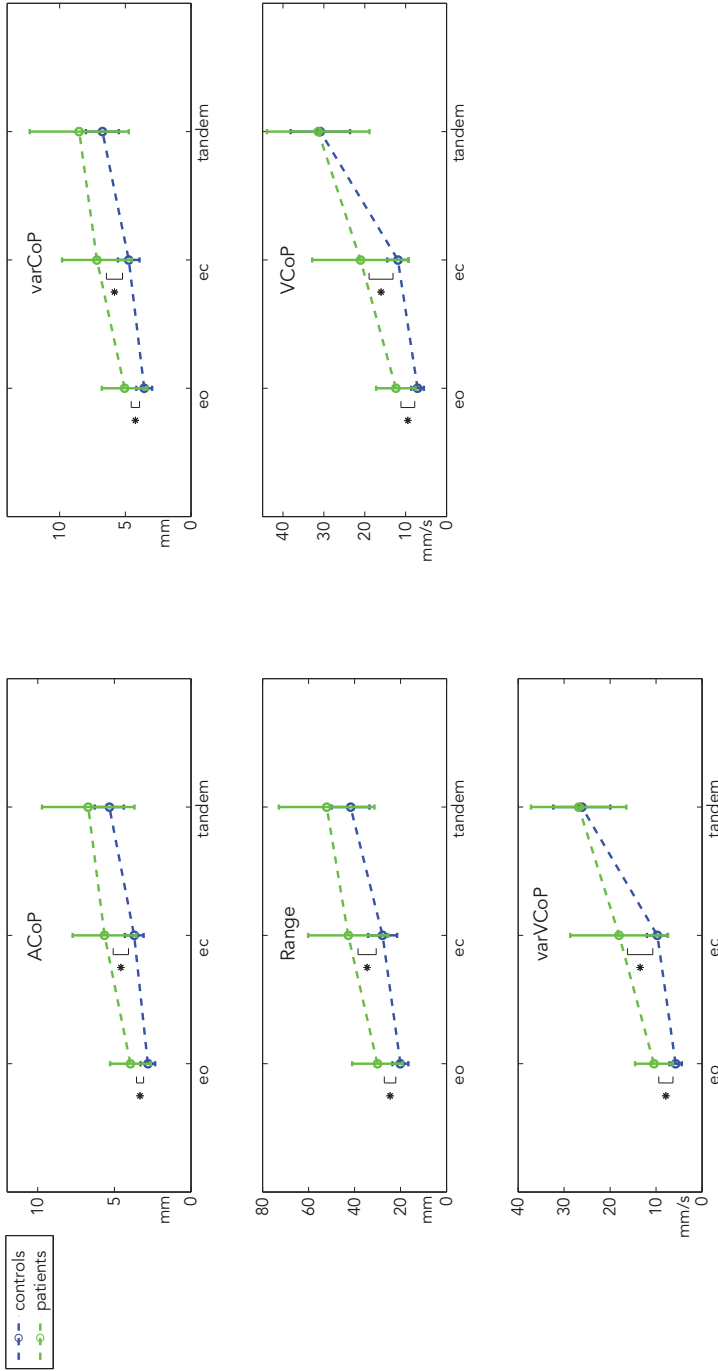
**Table 6.1** | Baseline characteristics and clinical assessments

Subjects' characteristics	Stroke subjects N=15	Healthy subjects N=10	P
Gender, male/female	12/3	6/4	0.29
Age in years (mean, SD)	57.1 (10.0)	57.9 (7.1)	0.82
Weight in kilograms (mean, SD)	86.1 (21.1)	78.2 (9.24)	0.41
Height in meters (mean, SD)	1.78 (0.10)	1.78 (0.73)	0.84
Time since stroke in months (mean, SD)	107.8 (143.6)	-	-
Affected hemisphere, right/left	9/6	-	-
Cortical/sub-cortical stroke	13/2	-	-
Bamford classification, LACI/PACI/TACI/unknown	7/4/2/2	-	-
Type of stroke, ischemic/haemorrhagic	11/4	-	-
CIRS, range 0–52 (median, IQR)	5 (4–6)	-	-
HADS, range 0–42 (median, IQR)	4 (3–10)	3.5 (0.75–5.25)	0.30
BBS, range 0–56 (median, IQR)	50 (48–53)	56 (56–56)	<0.01
TUG in seconds (mean, SD)	14.3 (7.9)	6.1 (0.99)	<0.01
EmNSA-LE, range 0–40 (median, IQR)	38 (34–39)	39 (39–40)	0.02
Falls past 6 months (median, IQR)	1 (0–2)	0.5 (0–1)	0.39
FES, range 7–28 (median, IQR)	10 (8–12)	-	-
FM-LE, range 0–34 (median, IQR)	25 (22–30)	-	-
MI-LE, range 0–100 (median, IQR)	69 (58–83)	-	-
MI-UE, range 0–100 (median, IQR)	84 (76–93.75)	-	-
Spatial neglect, yes/no	5/10	-	-

Overview of baseline characteristics and clinical assessments measured in the first session for 15 the patients and 10 healthy subjects. Mean per group are given as well as the standard deviation (SD) or the median and inter quartile ranges (IQR) in case of ordinal scales and the frequencies in case of nominal data. Deviation in cortical and sub-cortical lesions are made base on the main classification made by a clinician directly after stroke.

Abbreviations: total anterior circulation infarct (TACI), partial anterior circulation infarct (PACI), lacunar anterior circulation infarct (LACI), cumulative illness rating scale (CIRS), hospital anxiety and depression scale (HADS), Berg balance scale (BBS), timed up and go (TUG), Erasmus modification of the Nottingham sensory assessment lower extremity (EmNSA-LE), fall efficacy scale (FES), Fugl-Meyer assessment lower extremity (FM-LE), motricity index of the lower extremity (MI-LE) and the upper extremity (MI-UE), standardized score (Z-score), number per group (N), probability value (P).

Baseline CoP parameters



**Figure 6.3 |** Baseline CoP parameters. Baseline centre of pressure (CoP) parameters measured during the pre-stimulation eyes open (eo), eyes closed (ec) and the subject specific tandem stance (tandem) position. The mean amplitude of the CoP (ACoP), its amplitude's variability (varCoP), the velocity of the CoP (VCoP) and the velocity's variability (varVCoP) are displayed. Error bars indicate the standard deviation of the mean per group. \* Indicates a significant difference (probability value <0.05) between patients and healthy controls. Abbreviations: millimeters (mm), millimeters per second (mm/s).



### Effect of stimulation on CoP parameters

The tested model revealed no significant changes in CoP comp-score associated with cb-tDCS in the eyes open:  $\beta=0.02$  CI=-0.09–0.12,  $P=0.73$ , eyes closed:  $\beta=0.08$  CI=-0.01–0.16,  $P=0.07$  and tandem:  $\beta=-0.08$  CI=-0.41–0.25,  $P=0.64$  for the healthy controls. Adding the stimulation order to the model did not change  $\beta$ -values with more than 10%.

In the patient group, a significant association between contra-lesional stimulation and a decrease in CoP comp-score, in the tandem position was found:  $\beta=-0.25$ , CI=-0.48–0.03,  $P=0.03$ . Post-hoc analysis showed a significant decrease in ACoP:  $\beta=-0.86$ , CI=-1.58–0.15,  $P=0.02$ , varCoP:  $\beta=-1.10$ , CI=-1.93–0.26,  $P=0.01$ , range: ratio due to log transformation,  $\beta=0.94$ , CI=0.90–0.98,  $P=0.01$ , and VCOP: ratio due to log transformation,  $\beta=0.97$ , CI=0.94–0.99,  $P=0.02$  but not in varVCOP: ratio due to log transformation,  $\beta=0.97$ , CI=0.93–1.01,  $P=0.11$ . The GEE-model constructed for the eyes open position revealed a significant association between ipsi-lesional stimulation and a lower CoP comp-score:  $\beta=-0.09$ , CI=-0.18–0.01,  $P=0.03$ , after correcting for randomisation order, the association was no longer significant and changed to:  $\beta=0.00$ , CI=-0.09–0.90,  $P=0.94$  (Figures 6.4 and 6.5).

No changes in CoP comp-score in the eyes closed position associated with cb-tDCS were found for patients, contra-lesional stimulation:  $\beta=0.02$ , CI=-0.11–0.16,  $P=0.73$  and ipsi-lesional stimulation:  $\beta=-0.01$ , CI=-0.19–0.16,  $P=0.89$ . Adding the stimulation order to the model changed  $\beta$ -values with more than 10% to: contra-lesional stimulation:  $\beta=0.06$ , CI=-0.10–0.22,  $P=0.44$  and ipsi-lesional stimulation:  $\beta=0.09$ , CI=-0.08–0.26,  $P=0.30$ . See Table 6.2 for an overview of the corresponding  $\beta$ -values and confidence intervals.

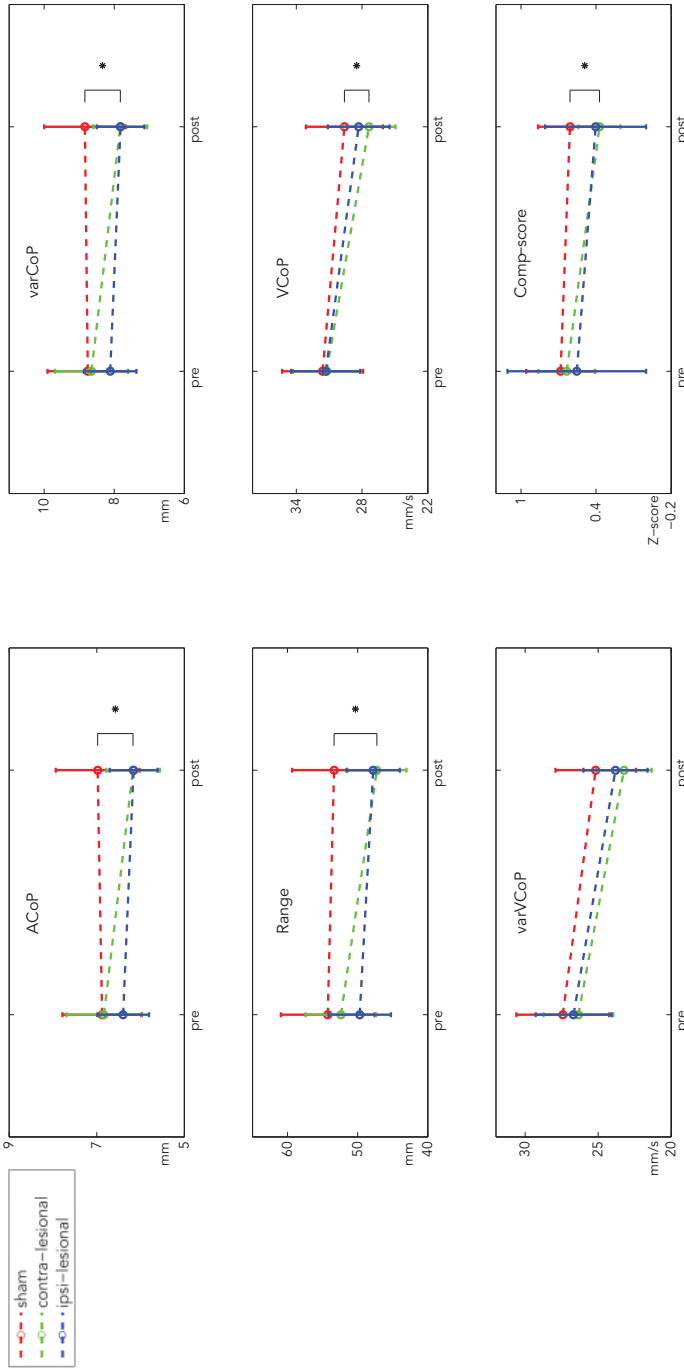
### Performance on the tracking task

The healthy controls showed a significantly higher tracking task performance during the second measurement; mean=99.33, SD=0.57,  $\beta=0.80$ , CI=0.10–1.50,  $P=0.03$  as compared to the first measurement; mean=98.53, SD=1.31. No effect of stimulation was found  $\beta=0$ , CI=-0.70–0.70,  $P=1.0$ .

The patient group showed a significantly higher tracking task performance during the second measurement; mean=92.90, SD=7.5, with a ratio of  $\beta=1.62$ , CI=1.23–2.14,  $P<0.01$  and on the third measurement; mean=94.60, SD=6.11, with a ratio of  $\beta=2.06$ , CI=1.51–2.80,  $P<0.01$  as compared to the first measurement; mean=88.60, SD=9.29.

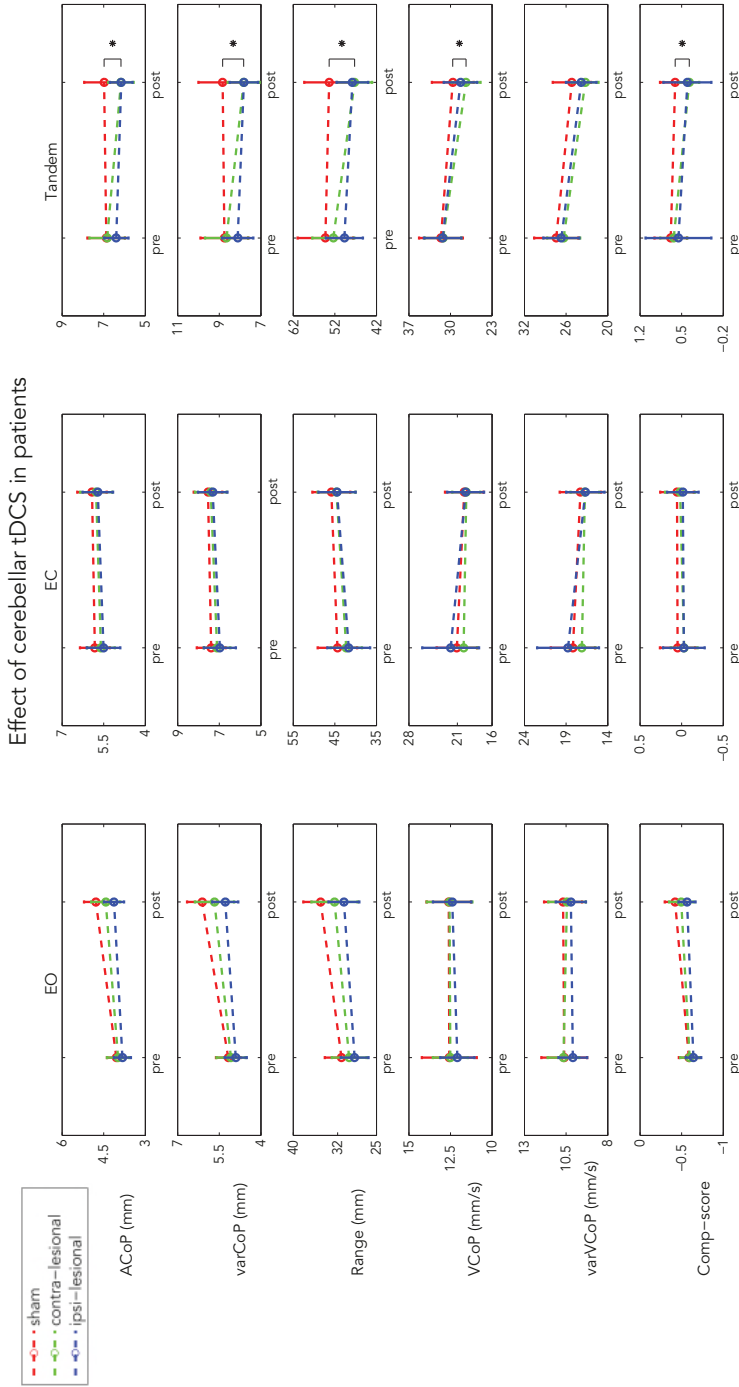
No effect of contra-lesional stimulation was found with a ratio due to log transformation of  $\beta=1.15$ , CI=0.89–1.50,  $P=0.29$ , nor an effect of ipsi-lesional stimulation,  $\beta=1.16$ , CI=0.91–1.49,  $P=0.23$ .

Effect of stimulation in patients, tandem stance



**Figure 6.4** | Effect of stimulation on tandem stance performance in stroke.

Centre of pressure (CoP) parameters measured during the pre-stimulation and post-stimulation in the subject specific tandem stance (tandem) positions. The mean amplitude of the CoP (ACoP), its amplitude's variability (varCoP), the velocity of the CoP (VCoP), the velocity's variability (varVCoP) and de composite-score are displayed. \* Indicates a significant difference with a probability value of <0.05, in the generalized estimating equation model with a correction for baseline and randomization order between contra-lesional cerebellar transcranial direct current stimulation (cb-tDCS) and the sham condition. Error bars indicate the standard error of the mean. Abbreviations: millimeters (mm), millimeters per second (mm/s).



**Figure 6.5** | CoP parameters measured pre and post stimulation in stroke patients. Centre of pressure (CoP) parameters measured during the pre-stimulation (pre) and post-stimulation (post) in the eyes open (eo), eyes closed (ec) and a subject specific tandem stance (tandem) position. The mean amplitude of the CoP (ACoP), its amplitude's variability (varCoP), the velocity of the CoP (VCoP), the velocity's variability (varVCoP) and de composite-score (Comp-score) are displayed. \* Indicates a significant difference (probability value <0.05) in the generalized estimating equation model with a correction for baseline and randomization order between contra-lesional cerebellar transcranial direct current stimulation (cb-tDCS) and sham. Error bars indicate the standard error of the mean. Note that the y-axis between conditions differ for visual inspection purposes. Abbreviations: millimeters (mm), millimeters per second (mm/s).

Table 6.2 | Overview of the tested models for the effect of stimulation

	Association of stimulation on a decrease in CoP Comp-score												
	Eyes open				Eyes closed				Tandem				
	$\beta$	95% CI	P		$\beta$	95% CI	P		$\beta$	95% CI	P		
<b>Controls (N=10)</b>													
Intercept	0.24	(-0.00-0.49)	0.05		-0.24	(-0.47-0.02)	0.04		0.31	(-0.83-0.70)	0.12		
Pre-stim score	1.17	(0.93-1.40)	0.00		-0.01	(-0.33-0.32)	0.96		0.80	(0.57-1.03)	0.00		
Stimulation	0.02	(-0.09-0.12)	0.73		0.08	(-0.01- 0.16)	0.07		-0.08	(-0.41-0.25)	0.64		
Intercept	0.24	(-0.01-0.49)	0.06		-0.27	(-0.47-0.06)	0.01		0.33	(-0.27-0.93)	0.28		
Pre-stim score	1.17	(0.93-1.40)	0.00		-0.05	(-0.47-0.30)	0.76		0.79	(0.53-1.05)	0.00		
Stimulation	0.02	(-0.08-0.12)	0.73		0.07	(-0.01-0.16)	0.07		-0.08	(-0.41-0.25)	0.64		
Order of measurements	0.01	(-0.08-0.10)	0.81		0.03	(-0.06-0.11)	0.57		-0.04	(-0.04-0.33)	0.84		
<b>Patients (N=15)</b>													
Intercept	0.13	(-0.05-0.30)	0.15		0.02	(-0.09-0.12)	0.75		0.16	(-0.04-0.36)	0.11		
Pre-stim score	0.93	(0.74-1.13)	0.00		0.76	(0.61-0.92)	0.00		0.65	(0.45-0.84)	0.00		
Ipsi-lesional stimulation	-0.09	(-0.18--0.01)	0.03		-0.01	(-0.19-0.16)	0.89		-0.12	(-0.30-0.06)	0.18		
Contra-lesional stimulation	-0.07	(-0.19-0.06)	0.32		0.02	(-0.11-0.16)	0.73		-0.25	(-0.48--0.03)	0.03		
Intercept	0.05	(-0.11- 0.21)	0.54		-0.04	(-0.20-0.12)	0.64		0.25	(0.07- 0.43)	0.01		
Pre-stim score	0.94	(0.79-1.09)	0.00		0.75	(0.60-0.91)	0.00		0.64	(0.46-0.82)	0.00		
Ipsi-lesional stimulation	0.00	(-0.09-0.90)	0.94		0.09	(-0.08-0.26)	0.30		-0.08	(-0.55-0.40)	0.75		
Contra-lesional stimulation	-0.02	(-0.14-0.01)	0.71		0.06	(-0.10-0.22)	0.44		-0.26	(-0.43--0.09)	0.00		
Sham at 3th session	-0.01	(-0.09-0.07)	0.78		-0.06	(-0.22-0.10)	0.44		-0.15	(-0.64-0.34)	0.54		
Sham at 2th session	0.12	(0.08-0.23)	0.04		0.08	(-0.08-0.24)	0.31		-0.11	(-0.29-0.06)	0.20		

Post-hoc analysis of the separate CoP parameters for the tandem position										
	ACoP			varCoP			Range*			
	$\beta$	95% CI	P	$\beta$	95%CI	P	$\beta$	95% CI	P	
Intercept	1.31	(0.16–2.46)	0.03	1.94	(0.37–3.51)	0.02	2.27	(1.40–3.66)	0.00	
Pre-stim score	0.82	(0.68–0.96)	0.00	0.80	(0.63–0.96)	0.00	1.26	(1.11–1.43)	0.00	
Ipsi-lesional stimulation	-0.31	(-1.37–0.75)	0.57	-0.30	(-1.74–1.13)	0.68	0.98	(0.89–1.06)	0.57	
Contra-lesional stimulation	-0.86	(-1.58–0.15)	0.02	-1.10	(-1.93–0.26)	0.01	0.94	(0.90–0.98)	0.01	
Sham at 3th session	-0.14	(-1.20–0.92)	0.80	-0.35	(-1.79–1.10)	0.64	0.97	(0.89–1.05)	0.44	
Sham at 2th session	0.01	(-0.72–0.75)	0.97	-0.11	(-0.97–0.76)	0.81	0.96	(0.91–0.99)	0.04	
	VCoP*			varVCoP*						
Intercept	1.38	(1.12–1.72)	0.00	1.63	(1.26–2.10)	0.00				
Pre-stim score	1.40	(1.30–1.50)	0.00	1.32	(1.21–1.44)	0.00				
Ipsi-lesional stimulation	0.98	(0.87–1.1)	0.68	0.98	(0.85–1.12)	0.73				
Contra-lesional stimulation	0.97	(0.94–0.99)	0.02	0.97	(0.93–1.01)	0.11				
Sham at 3th session	1.00	(0.89–1.12)	0.96	0.98	(0.86–1.12)	0.78				
Sham at 2th session	0.98	(0.95–1.00)	0.07	0.96	(0.92–1.01)	0.08				

Overview of the generalized estimating equation models that were tested for the effect of stimulation on post-measurement center of pressure (CoP) parameters in healthy controls and patients. All models were corrected for the individual pre-measurement CoP scores. If the  $\beta$ -values in the model changed more than 10% by correcting for the randomized order of the stimulation to the model, this was considered an improvement of the model. Sham stimulation condition or sham stimulation condition at the first session was used as contrast.

Parameters marked with an asterisk (\*) were natural logarithmic transformed, the exponential functions of  $\beta$  and the CI are given in the table and should be interpreted as ratios. Abbreviations: the mean amplitude of the CoP (ACoP), its amplitude's variability (varCoP), the velocity of the CoP (VCoP), the velocity's variability (varVCoP), Unstandardized Beta-value ( $\beta$ ), confidence interval (CI), probability value (P).

## Responders and non-responders

Ten patients had a reduction in CoP comp-score in the tandem stance in response to contra-lesional stimulation (responders). Five patients did not show a change when compared to sham stimulation (non-responders). The group responders did not differ on any of the baseline CoP comp-scores, fatigue level or clinical characteristics from the non-responders, see Table 6.3. Out of the 5 non-responders, 2 patients could be identified as responders on the ipsi-lesional stimulation, while 5 patients responded to both stimulation types.

**Table 6.3** | Differences in characteristics and clinical assessments between responders and non-responders

Subject characteristics	Responders N=10	Non-responders N=5	P
Age in years (mean, SD)	54.4 ( $\pm 8.88$ )	62.4 ( $\pm 10.94$ )	0.15
Time since stroke in months (mean, SD)	76.9 ( $\pm 86.2$ )	169.8 ( $\pm 219.8$ )	0.25
Affected hemisphere, right/left	5/5	4/1	0.18
Bamford classification, LACI/PACI/TACI/unknown	4/3/1/2	3/1/1/0	0.23
Type of stroke, ischemic/haemorrhagic	9/1	2/3	0.07
CIRS, range 0–52 (median, IQR)	5 (3.75–6.25)	5 (3.5–7)	1
HADS, range 0–42 (median, IQR)	4 (3–10.25)	5 (1.5–8)	0.85
BBS, range 0–56 (median, IQR)	50 (48.75–52.25)	52 (43–53.5)	0.76
TUG in seconds (mean, SD)	11 (8.75–17)	15 (8–20.5)	0.67
EmNSA-LE, range 0–40 (median, IQR)	38 (35.5–39.25)	37 (30.5–39)	0.46
FES, range 7–28 (median, IQR)	10 (7.75–12.5)	11 (7.5–14)	0.95
Falls past 6 months (median, IQR)	1 (0–1.5)	2 (0–2)	0.70
MI-LE, range 0–100 (median, IQR)	26 (22.75–30.25)	24 (13–28)	0.30
MI-UE, range 0–100 (median, IQR)	70.5 (62.5–85)	59 (45.5–72)	0.16
Spatial neglect, yes/no	3/7	2/3	0.20
Fatigue VAS, range 0–100 (median, IQR)	2.2 ( $\pm 3.2$ )	1.6 ( $\pm 1.2$ )	0.70
CoP pre_comp_EO (mean z-score, SD)	-0.43 ( $\pm 0.56$ )	-0.79 ( $\pm 0.28$ )	0.20
CoP pre_comp_EC (mean z-score, SD)	0.04 ( $\pm 0.61$ )	-0.15 ( $\pm 0.74$ )	0.63
CoP pre_comp_tandem (mean z-score, SD)	0.83 ( $\pm 1.23$ )	0.32 ( $\pm 0.52$ )	0.39

Overview of characteristics and clinical assessments measured in the first session for 10 responders and 5 non-responders on the contra-lesional cb-tDCS. The assessment range is given in case of ordinal scales. Mean per group are given as well as the standard deviation (SD) or the median and inter quartile ranges (IQR) in case of ordinal scales and the frequencies in case of nominal data.

Abbreviations: total anterior circulation infarct (TACI), partial anterior circulation infarct (PACI), lacunar anterior circulation infarct (LACI), cumulative illness rating scale (CIRS), hospital anxiety and depression scale (HADS), Berg balance scale (BBS), timed up and go (TUG), Erasmus modification of the Nottingham sensory assessment lower extremity (EmNSA-LE), fall efficacy scale (FES), Fugl-Meyer assessment lower extremity (FM-LE), motricity index of the lower extremity (MI-LE), visual analog scale (VAS), first measured session of the centre of pressure composite score (CoP pre\_comp) for eyes open (EO), eyes closed (EC) and the tandem stance (tandem) position, standardized score (Z-score), Number per group (N), probability value (P).

## DISCUSSION

To our knowledge this is the first study reporting the short-term effect of cb-tDCS on standing balance performance in patients with chronic stroke. The effect of cb-tDCS was tested during three static positions, namely: eyes open, eyes closed and in a tandem position. No effects of cb-tDCS on standing balance performance were found in the first two positions for patients nor in control subjects. In the tandem stance position a significant decrease in four separate CoP parameters and in the CoP comp-score was found, suggesting an improvement in standing balance performance in the stroke patients after contra-lesional anodal stimulation. In healthy controls no effect of cb-tDCS was found in the tandem stance position.

tDCS is believed to facilitate motor learning while being simultaneously applied with a motor task<sup>41</sup>. It could therefore be expected that the most difficult task, the (semi-)tandem position in which there is most to gain, would show the largest improvement in standing balance performance after training with cb-tDCS. It is likely that the tandem stance position and the tracking task were not difficult enough for the healthy controls. Similar to the study by Steiner et al.<sup>25</sup>, who used a moving platform to train balance performance in 30 healthy young adults and found no effects of cb-tDCS, healthy subjects in our study were also likely performing on a (sub)optimal level with a mean performance of 98.5% after the first session and had very little room for improvement. Future studies with an interest in the feasibility of cb-tDCS in healthy subjects, should use a more challenging postural task or a dual task paradigm.

Poortvliet et al.<sup>23</sup> found a significant smaller CoP path length and standard deviation during and after disturbed proprioceptive input with Achilles tendon vibration in quiet stance, in the group receiving cb-tDCS as compared to sham stimulation. This is an interesting model in the understanding of cb-tDCS on balance performance, since dysfunction of proprioception can occur after stroke and can hamper balance performance<sup>42</sup>. It would be interesting to study whether proprioceptive function is an independent covariate for improvement of balance performance after cb-tDCS in patients after stroke. Unfortunately the sample size in the current study was too small to perform such a sub-analysis. The reported changes in standing balance performance in the tandem stance position with contra-lesional stimulation in patients with a stroke, are in line with the theoretical framework that both LTP and LTD like processes play a role in spike timing-dependent forms of neuroplasticity in the cerebellum and non-invasive stimulation may enhance these processes<sup>43</sup>. Theoretically, stimulation on either one of the cerebellar hemispheres could enhance adaptive motor learning and thereby improve motor coordination.



Targeting the contra-lesional cerebellar hemisphere could directly strengthen the M1-cerebellar connection, to enhance the function of the cerebellum connected to the affected cortical hemisphere<sup>16,24</sup>. On the other hand, neural activity of the ipsilateral cortex during movements has been associated with poor functional outcome after stroke<sup>44</sup>. This phenomenon has been attributed to a decreased inter hemispheric inhibition (IHI) from the lesioned motor cortex on the non-affected hemisphere, leading to an increased IHI on the affected hemisphere, negatively affecting functional outcome<sup>45</sup>. Inhibition of the ipsilateral motor cortex with cathodal stimulation is thought to suppress this over activity<sup>46</sup>. Anodal stimulation of the ipsi-lesional cerebellar hemisphere could have potentially improved the disturbed IHI in an indirect manner via cerebellar brain inhibition and at the same time enhance adaptive learning. The results of this study did not show evidence that targeting IHI via the cerebellum can lead to improvement in terms of standing balance performance. From these results it cannot yet be concluded if the protocol failed to induce a normalisation in IHI or if it should be considered a compensatory mechanism reflecting the severity of the brain damage, in which normalization of this phenomena does not have any added value for clinical outcome<sup>47</sup>. Future research should aim to underpin the neurobiological mechanisms by which cb-tDCS enforces its effects.

### **Baseline differences between patients and healthy controls**

The increase in postural sway found in stroke patients compared to aged match controls and the enlarged sway with the more difficult positions, are in line with previous found results in a comparable population<sup>48</sup>. However, no significant differences in CoP parameters were found between stroke patients and controls for the tandem stance position. This result may be explained by the individualized feet positioning during the tandem stance per subject. None of the stroke patients could hold the full tandem stance position with either leg behind, while all but one healthy subject could hold this position with the non-preferred leg behind. This again indicates that the task may not have been difficult enough for the healthy controls. The main purpose of this proof of concept study was to investigate the potential of cb-tDCS to elicit qualitative changes in standing balance performance in patients with a stroke. Since the differences between patients are much larger than the effect that can be expected from a single training session, a within subject design was needed with a challenging task, tailored to the specific capacity of each patient.

### **Outcome parameters**

A decrease in CoP parameters is generally assumed to reflect an improvement in postural stability in patients with a stroke<sup>49</sup>. There is a strong need for more sensitive and reliable

measures to be able to quantify subtle changes in standing balance performance and disentangle postural control mechanisms<sup>50</sup>. Despite several promising methods to quantify postural control, a golden standard is still lacking<sup>51,52</sup>. We used the CoP comp-score as a sensitive comprehensive outcome parameter, combining information from five parameters of standing balance performance<sup>40</sup>. A sensitive outcome parameter, able to detect subtle qualitative changes, is also needed to establish an optimal dose in terms of sessions and intensity. To relate the currently found short term effects of anodal cb-tDCS on standing balance performance to a clinical meaningful and long-term improvement, the effect of multiple training sessions should be measured using both qualitative parameters of standing balance performance and clinical outcome measures.

### Responders and non-responders

A general point of concern in tDCS research is the different responsiveness of subjects to the stimulation, i.e. why do some subjects respond to the stimulation while others don't? Several reviews on both healthy subjects as well as patients with stroke have highlighted factors such as: age, anxiety, time since stroke, lesion type, lesion location and motor function, as contributors to interindividual variability in response to tDCS<sup>53-57</sup>. Within this study ten patients showed changes towards normal CoP values in response to contralesional cb-tDCS compared to sham. On a group level this contrast was large enough for a significant association, the non-responders should however not be ignored. These interindividual differences play a role in many tDCS studies, which lead to ambiguity in the interpretation of results and the conclusion of some authors that tDCS does not have any added value or vice versa<sup>58</sup>. Within the small sample of this study no differences could be detected between responders and non-responders on several subject characteristics. Next to clinical characteristics, the initial state of neuronal populations could play a role in terms of responsiveness to the stimulation<sup>55,59</sup>. Neuroimaging techniques could provide valuable insights into these neuronal state dependencies<sup>60</sup>.

### Strength and limitations

This proof of concept study is the first aimed at the short-term effect on standing balance performance of a single training sessions combined with anodal cb-tDCS in stroke patients. The current setup was able to detect subtle qualitative changes in standing balance performance by using high resolution kinetic parameters. The study population was however small and heterogeneity in terms of lesion type, location and motor function could have influenced the interindividual variability in response to the stimulation. Within the sample 2 patients were included with a lesion primarily located in the brainstem,

both of these patients were classified as responders to the contra-lesional cb-tDCS. The current sample is however too small to perform a sub-group analysis or to generalize these findings to a wider population. The healthy controls in this study only received stimulation ipsi-lateral to their dominant leg. It is possible that an improvement in the healthy subjects could have been found after contralateral stimulation. The more evident explanation for the lack of improvement in healthy controls is however that the task was too easy and a more challenging task should have been chosen for this group.

### **Future research**

The current study shows the potential of anodal cb-tDCS for improving standing balance performance in a chronic stroke population, though interindividual differences should be further studied. Analysis of ongoing cortical processes during standing balance tasks and the influence of cb-tDCS on these processes may give more insights in unknown underlying mechanisms. Moreover, these unknown mechanisms leading to interindividual differences, could be very different in the more acute phase after stroke. Behavioural restitution of function, mainly taking place in the first 8–12 weeks post-stroke <sup>61</sup>, goes alongside changes in growth factors, creating a critical time window for recovery <sup>62–64</sup>. tDCS might be able to optimize the learning potential in this time window, leading to a completely different paradigm of the mechanisms of action of tDCS in the sub-acute phase than in a population of patients in a chronic phase after stroke <sup>65,66</sup>. Considering that balance recovery is especially important in the early phase after stroke, tDCS interventions should also be tested in this time window of enhanced neuroplasticity. In addition, possible contributors to interindividual differences, such as a wide variety of clinical characteristics as well as neuronal state, should be recorded.

## **CONCLUSION**

The improvement in standing balance performance after anodal contra-lesional cerebellar tDCS shows promise for the application in stroke rehabilitation. Future studies should investigate interindividual differences to elucidate the working mechanisms of tDCS. Qualitative outcome parameters that can capture the subtle effects of tDCS can be used to explore the optimal dose, and should be related to clinically meaningful improvements. High quality randomized controlled trials in the early phase after stroke are needed to establish the role of cb-tDCS in the critical time window of recovery post-stroke and its potential to enhance clinical outcome in rehabilitation practice.

## Funding

This study was funded by a fellowship of the Dutch brain foundation (Hersenstichting), the Netherlands, awarded to dr. E.E.H. van Wegen. Grant nr: F2013(1)-41.

## REFERENCES

- 1 Geurts ACH, De Haart M, Van Nes IJW, Duysens J. A review of standing balance recovery from stroke. *Gait Posture* 2005; **22**: 267–281.
- 2 Veerbeek J, Van Wegen E, Van Peppen R, Van Der Wees PJ, Hendriks E, Rietberg M *et al.* What is the evidence for physical therapy poststroke? A systematic review and meta-analysis. *PLoS One* 2014; **9**: e87987.
- 3 Reis J, Fritsch B. Modulation of motor performance and motor learning by transcranial direct current stimulation. *Curr Opin Neurol* 2011; **24**: 590–596.
- 4 Stagg CJ, Jayaram G, Pastor D, Kincses ZT, Matthews PM, Johansen-Berg H. Polarity and timing-dependent effects of transcranial direct current stimulation in explicit motor learning. *Neuropsychologia* 2011; **49**: 800–804.
- 5 Marquez J, van Vliet P, Mcelduff P, Lagopoulos J, Parsons M. Transcranial direct current stimulation (tDCS): Does it have merit in stroke rehabilitation? A systematic review. *Int J Stroke* 2015; **10**: 306–316.
- 6 Kwakkel G, Lannin NA, Borschmann K, English C, Ali M, Churilov L *et al.* Standardized measurement of sensorimotor recovery in stroke trials: Consensus-based core recommendations from the Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2017; **12**: 451–461.
- 7 Bernhardt J, Kwakkel G, Lannin NA, Borschmann K, English C, Ali M *et al.* Consensus Statements from the Stroke Recovery and Rehabilitation Roundtable Standardized Measurement of Sensorimotor Recovery in Stroke Trials: Consensus-Based Core Recommendations from the Stroke Recovery and Rehabilitation Roundtable\*. *Int J Stroke* 2017; **31**: 784–792.
- 8 Cooke SF, Bliss TVP. Plasticity in the human central nervous system. *Brain* 2006; **129**: 1659–1673.
- 9 Caligiore D, Pezzulo G, Baldassarre G, Bostan AC, Strick PL, Doya K *et al.* Consensus Paper: Towards a Systems-Level View of Cerebellar Function: the Interplay Between Cerebellum, Basal Ganglia, and Cortex. *Cerebellum* 2017; **16**: 203–229.
- 10 Jayaram G, Galea JM, Bastian AJ, Celnik P. Human locomotor adaptive learning is proportional to depression of cerebellar excitability. *Cereb Cortex* 2011; **21**: 1901–1909.
- 11 Medina JF, Lisberger SG. Links from complex spikes to local plasticity and motor learning in the cerebellum of awake-behaving monkeys. *Nat Neurosci* 2008; **11**: 1185–1192.
- 12 Ito M, Yamaguchi K, Nagao S, Yamazaki T. Long-Term Depression as a Model of Cerebellar Plasticity. *Prog Brain Res* 2014; **210**: 1–30.
- 13 Morton SM, Bastian AJ. Cerebellar control of balance and locomotion. *Neuroscientist* 2004; **10**: 247–259.
- 14 Pollok B, Butz M, Gross J, Südmeyer M, Timmermann L, Schnitzler A. Coupling between cerebellar hemispheres: Behavioural, anatomic, and functional data. *Cerebellum* 2006; **5**: 212–219.
- 15 Galea JM, Vazquez A, Pasricha N, Orban De Xivry JJ, Celnik P. Dissociating the roles of the cerebellum and motor cortex during adaptive learning: The motor cortex retains what the cerebellum learns. *Cereb Cortex* 2011; **21**: 1761–1770.
- 16 Schlerf JE, Galea JM, Spampinato D, Celnik PA. Laterality Differences in Cerebellar-Motor Cortex Connectivity. *Cereb Cortex* 2015; **25**: 1827–1834.
- 17 Akaike T. Neuronal organization of the vestibulospinal system in the cat. *Brain Res* 1983; **259**: 217–227.
- 18 Rampersad SM, Janssen AM, Lucka F, Aydin U, Lanfer B, Lew S *et al.* Simulating transcranial direct current stimulation with a detailed anisotropic human head model. *IEEE Trans Neural Syst Rehabil Eng* 2014; **22**: 441–452.
- 19 Nitsche M a, Paulus W. Excitability changes induced in the human motor cortex by weak transcranial direct current stimulation. *J Physiol* 2000; **527**: 633–639.
- 20 Galea JM, Jayaram G, Ajagbe L, Celnik P. Modulation of cerebellar excitability by polarity-specific noninvasive direct current stimulation. *J Neurosci* 2009; **29**: 9115–9122.
- 21 Inukai Y, Saito K, Sasaki R, Kotan S, Nakagawa M, Onishi H. Influence of Transcranial Direct Current Stimulation to the Cerebellum on Standing Posture Control. *Front Hum Neurosci* 2016; **10**: 325.

- 22 Foerster Á, Melo L, Mello M, Castro R, Shirahige L, Rocha S *et al.* Cerebellar Transcranial Direct Current Stimulation (ctDCS) Impairs Balance Control in Healthy Individuals. *Cerebellum* 2017; **16**: 872–875.
- 23 Poortvliet P, Hsieh B, Cresswell A, Au J, Meinzer M. Cerebellar transcranial direct current stimulation improves adaptive postural control. *Clin Neurophysiol* 2018; **129**: 33–41.
- 24 Jayaram G, Tang B, Pallegadda R, Vasudevan EVL, Celnik P, Bastian A. Modulating locomotor adaptation with cerebellar stimulation. *J Neurophysiol* 2012; **107**: 2950–2957.
- 25 Steiner KM, Enders A, Thier W, Batsikadze G, Ludolph N, Ilg W *et al.* Cerebellar tDCS Does Not Improve Learning in a Complex Whole Body Dynamic Balance Task in Young Healthy Subjects. *PLoS One* 2016; **11**: e0163598.
- 26 Schonewille M, Belmeguenai A, Koekoek SK, Houtman SH, Boele HJ, van Beugen BJ *et al.* Purkinje cell-specific knockout of the protein phosphatase PP2B impairs potentiation and cerebellar motor learning. *Neuron* 2010; **67**: 618–628.
- 27 Schonewille M, Gao Z, Boele HJ, Vinueza Veloz MF, Amerika WE, Simek AAM *et al.* Reevaluating the Role of LTD in Cerebellar Motor Learning. *Neuron* 2011; **70**: 43–50.
- 28 D'Angelo E. The Organization of Plasticity in the Cerebellar Cortex: From Synapses to Control. *Prog Brain Res* 2014; **210**: 31–58.
- 29 Zigmund AS, Snaith RP. The hospital anxiety and depression scale (HADS). *Acta Psychiatr Scand* 1983; **67**: 361–370.
- 30 Poreisz C, Boros K, Antal A, Paulus W. Safety aspects of transcranial direct current stimulation concerning healthy subjects and patients. *Brain Res Bull* 2007; **72**: 208–214.
- 31 Blum L, Korner-Bitensky N. Usefulness of the Berg Balance Scale in stroke rehabilitation: a systematic review. *Phys Ther* 2008; **88**: 559–566.
- 32 Duncan PW, Weiner DK, Chandler J, Studenski S. Functional reach: a new clinical measure of balance. *J Gerontol* 1990; **45**: 192–197.
- 33 Shumway-Cook A, Brauer S, Woollacott M. Predicting the probability for falls in community-dwelling older adults using the Timed Up & Go Test. *Phys Ther* 2000; **80**: 896–903.
- 34 Tinetti ME, Richman D, Powell L. Falls efficacy as a measure of fear of falling. *J Gerontol* 1990; **45**: 239–243.
- 35 Stolk-Hornsveld F, Crow JL, Hendriks EP, Van Der Baan R, Harmeling-van Der Wel BC. The Erasmus MC modifications to the (revised) Nottingham Sensory Assessment: a reliable somatosensory assessment measure for patients with intracranial disorders. *Clin Rehabil* 2006; **20**: 160–172.
- 36 Duncan PW, Propst M, Nelson SG. Reliability of the Fugl-Meyer assessment of sensorimotor recovery following cerebrovascular accident. *Phys Ther* 1983; **63**: 1606–1610.
- 37 Collin C, Wade D. Assessing motor impairment after stroke: a pilot reliability study. *J Neurol Neurosurg Psychiatry* 1990; **53**: 576–579.
- 38 Harwood RH, Ebrahim S. The validity, reliability and responsiveness of the Nottingham Extended Activities of Daily Living scale in patients undergoing total hip replacement. *Disabil Rehabil* 2002; **24**: 371–377.
- 39 Ferber S, Karnath HO. How to assess spatial neglect—line bisection or cancellation tasks? *J Clin Exp Neuropsychol* 2001; **23**: 599–607.
- 40 Pasma JH, Bijlsma AY, Van Der Bij MDW, Arendzen JH, Meskers CGM, Maier AB. Age-related differences in quality of standing balance using a composite score. *Gerontology* 2014; **60**: 306–314.
- 41 Nitsche M a, Schauenburg A, Lang N, Liebetanz D, Exner C, Paulus W *et al.* Facilitation of implicit motor learning by weak transcranial direct current stimulation of the primary motor cortex in the human. *J Cogn Neurosci* 2003; **15**: 619–626.
- 42 Bonan I V, Colle FM, Guichard JP, Vicaud E, Eisenfisz M, Tran Ba Huy P *et al.* Reliance on visual information after stroke. Part I: Balance on dynamic posturography. *Arch Phys Med Rehabil* 2004; **85**: 268–273.
- 43 Cooke SF, Bliss TVP. Plasticity in the human central nervous system. *Brain* 2006; **129**: 1659–73.
- 44 Ward NS, Brown MM, Thompson AJ, Frackowiak RSJ. Neural correlates of outcome after stroke: a cross-sectional fMRI study. *Brain* 2003; **126**: 1430–1448.

- 45 Murase N, Duque J, Mazzocchio R, Cohen LG. Influence of Interhemispheric Interactions on Motor Function in Chronic Stroke. *Ann Neurol* 2004; **55**: 400–409.
- 46 Di Lazzaro V, Dileone M, Capone F, Pellegrino G, Ranieri F, Musumeci G et al. Immediate and late modulation of interhemispheric imbalance with bilateral transcranial direct current stimulation in acute stroke. *Brain Stimul* 2014; **7**: 841–848.
- 47 Di Pino G, Pellegrino G, Assenza G, Capone F, Ferreri F, Formica D et al. Modulation of brain plasticity in stroke: a novel model for neurorehabilitation. *Nat Rev Neurol* 2014; **10**: 597–608.
- 48 de Haart M, Geurts AC, Huidekoper SC, Fasotti L, van Limbeek J. Recovery of standing balance in postacute stroke patients: a rehabilitation cohort study. *Arch Phys Med Rehabil* 2004; **85**: 886–895.
- 49 Genthon N, Rougier P, Gissot A-S, Froger J, Pelissier J, Perennou D. Contribution of each lower limb to upright standing in stroke patients. *Stroke* 2008; **39**: 1793–1799.
- 50 Meskers CGM, de Groot JH, de Vlugt E, Schouten AC. NeuroControl of movement: system identification approach for clinical benefit. *Front Integr Neurosci* 2015; **9**: 48.
- 51 Pasma JH, Engelhart D, Maier AB, Schouten AC, van der Kooij H, Meskers CGM et al. Changes in sensory reweighting of proprioceptive information during standing balance with age and disease. *J Neurophysiol* 2015; **114**: 3220–3233.
- 52 de Kam D, Kamphuis JF, Weerdesteyn V, Geurts ACH. The effect of weight-bearing asymmetry on dynamic postural stability in people with chronic stroke. *Gait Posture* 2017; **53**: 5–10.
- 53 de Aguiar V, Paolazzi CL, Miceli G. tDCS in post-stroke aphasia: The role of stimulation parameters, behavioral treatment and patient characteristics. *Cortex* 2015; **63**: 296–316.
- 54 Lefebvre S, Liew S-L. Anatomical Parameters of tDCS to Modulate the Motor System after Stroke: A Review. *Front Neurol* 2017; **8**: 29.
- 55 Wiethoff S, Hamada M, Rothwell JC. Variability in response to transcranial direct current stimulation of the motor cortex. *Brain Stimul* 2014; **7**: 468–475.
- 56 Hsu T-Y, Juan C-H, Tseng P. Individual Differences and State-Dependent Responses in Transcranial Direct Current Stimulation. *Front Hum Neurosci* 2016; **10**: 643.
- 57 Li LM, Uehara K, Hanakawa T. The contribution of interindividual factors to variability of response in transcranial direct current stimulation studies. *Front Cell Neurosci* 2015; **9**: 181.
- 58 Horvath JC, Forte JD, Carter O. Evidence that transcranial direct current stimulation (tDCS) generates little-to-no reliable neurophysiologic effect beyond MEP amplitude modulation in healthy human subjects: A systematic review. *Neuropsychologia* 2015; **66**: 213–236.
- 59 Silvanto J, Muggleton N, Walsh V. State-dependency in brain stimulation studies of perception and cognition. *Trends Cogn Sci* 2008; **12**: 447–454.
- 60 Al-Kaysi AM, Al-Ani A, Loo CK, Breakspear M, Boonstra TW. Predicting brain stimulation treatment outcomes of depressed patients through the classification of EEG oscillations. *2016 38th Annu Int Conf IEEE Eng Med Biol Soc* 2016; **5266**–5269.
- 61 Kwakkel G, Kollen B, Twisk J. Impact of time on improvement of outcome after stroke. *Stroke* 2006; **37**: 2348–2353.
- 62 Bernhardt J, Borschmann K, Boyd L, Carmichael ST, Corbett D, Cramer SC et al. Moving Rehabilitation Research Forward: Developing Consensus Statements for Rehabilitation and Recovery Research. *Int J Stroke* 2016; **11**: 454–458.
- 63 Cramer SC. Repairing the human brain after stroke: I. Mechanisms of spontaneous recovery. *Ann Neurol* 2008; **63**: 272–287.
- 64 Murphy TH, Corbett D. Plasticity during stroke recovery: from synapse to behaviour. *Nat Rev Neurosci* 2009; **10**: 861–872.
- 65 Krakauer JW, Carmichael ST, Corbett D, Wittenberg GF. Getting Neurorehabilitation Right: What Can Be Learned From Animal Models? *Neurorehabil Neural Repair* 2012; **26**: 923–931.
- 66 Sattler V, Acket B, Raposo N, Albuchoer JF, Thalamas C, Loubinoux I et al. Anodal tDCS Combined with Radial Nerve Stimulation Promotes Hand Motor Recovery in the Acute Phase after Ischemic Stroke. *Neurorehabil Neural Repair* 2015; **29**: 743–754.









# Chapter 7

The effect of cerebellar transcranial direct current stimulation to improve standing balance performance early post-stroke, study protocol of a randomised controlled trial

Sarah B. Zandvliet  
Carel G.M. Meskers  
Rinske H.M. Nijland  
Andreas Daffertshofer  
Gert Kwakkel  
Erwin E.H. van Wegen

*International Journal of Stroke* 2019;  
14(6): 650–657

## ABSTRACT

**Rationale:** Restoration of adequate standing balance after stroke is of major importance for functional recovery. POstural feedback ThErapy combined with Non-invasive TranscranIAL direct current stimulation (tDCS) in patients with stroke (acronym POTENTIAL) aims to establish if cerebellar tDCS has added value in improving standing balance performance early post-stroke.

**Methods:** Forty-six patients with a first-ever ischemic stroke will be enrolled in this double-blind controlled trial within 5 weeks post-stroke. All patients will receive fifteen sessions of virtual reality based postural feedback training (VR-PFT) in addition to usual care. VR-PFT will be given 5 days per week for 1 hour, starting within 5 weeks post-stroke. During VR-PFT, 23 patients will receive 25 minutes of cerebellar anodal tDCS (cb-tDCS), and 23 patients will receive sham stimulation.

**Study outcome:** Clinical, posturographic and neurophysiological measurements will be performed at baseline, directly post-intervention, two weeks post-intervention and at fifteen weeks post-stroke. The primary outcome measure will be the Berg balance scale (BBS) for which a clinical meaningful difference of 6 points needs to be established between the intervention and control group at fifteen weeks post-stroke.

**Discussion:** POTENTIAL will be the first proof-of-concept randomised controlled trial to assess the effects of VR-PFT combined with cerebellar tDCS in terms of standing balance performance in patients early post-stroke. Due to the combined clinical, posturographical and neurophysiological measurements, this trial may give more insights in underlying post-stroke recovery processes and whether these can be influenced by tDCS.



## INTRODUCTION AND RATIONALE

Impaired standing balance after stroke is common and has a significant impact on fall events, independence in activities of daily living and perceived disability<sup>1,2</sup>. Prospective cohort studies suggest that most improvements in standing balance and walking ability occur within the first 5 to 8 weeks post-stroke<sup>3,4</sup>. There is strong evidence of enhanced homeostatic forms of neuroplasticity during this time window, including upregulation of gene expression of growth promoting factors, such as brain derived nerve growth factors (BDNF) followed by growth inhibiting factors<sup>5</sup>. Human motor learning in this critical time window may be facilitated by transcranial direct current stimulation (tDCS) which is believed to specifically target synapse-based learning by enhancing the turnover of the secretion of BDNF<sup>6</sup>. tDCS is thought to induce polarity-driven alterations of membrane potentials and efficacy modulations of specific neuronal receptors in the underlying brain tissue<sup>6</sup>.

These dynamic neural modulations are evident not only in motor performance<sup>7-9</sup>, but also in intrinsic functional network connectivity that manifest in neurophysiological recordings of cortical brain activity<sup>10</sup>. Neural changes while performing balance tasks are mostly reflected by a change in theta (4–8 Hz), and alpha power (8–12.5 Hz)<sup>11,12</sup>. A higher alpha power reflects increased learning speed and an optimal concentration level<sup>13</sup>. Decreased alpha activity is also generally seen in patients after stroke<sup>14</sup>. Theta power activity is associated with an emerging state of concentration and optimal error control and found to increase with increasing complexity of balance tasks<sup>11,15</sup>. Although a general deceleration of EEG signals is associated with poor functional outcome after stroke, conflicting results regarding a correlation of increased theta power activity with post-stroke function are found<sup>16-18</sup>. Next to an alteration in power spectral density, asymmetry between the hemispheres (low brain symmetry index) has been associated with poor clinical function and disability 6 months post stroke and is believed to reflect the clinical neurological condition of acute stroke patients<sup>19,20</sup>. To study these changes in cortical activation patterns in post-stroke recovery, and the potential influence tDCS may have on these processes, repetitive EEG measurements in both a resting state and during postural balance tasks are required<sup>21-23</sup>.

The cerebellum with its distinct role in feedback based learning could be a promising target for tDCS<sup>8,24</sup>. The cerebellum is involved in motor adaptation via long term depression (LTD)-like plasticity of Purkinje cells mediated by activation of predominantly climbing fibers<sup>25-27</sup>. Via cortico-cerebellar connections, it is involved in optimisation of timing of movements by comparing a copy of efferent and afferent information, which may be enhanced by tDCS<sup>28-31</sup>. From a detailed anisotropic head model study a known optimal configuration to apply cerebellar tDCS is available<sup>32</sup>.

We recently found an instantaneous positive effect of a postural feedback based tracking task combined with anodal cerebellar tDCS (cb-tDCS) on standing balance performance in a small group of patients with a chronic stroke (N=15) when stimulated on the ipsi-lesional cerebellar hemisphere as compared to sham <sup>33</sup>. Moreover, it has been proposed that anodal cb-tDCS may counteract the effect of crossed cerebellar diaschisis, which induces a disbalance in cerebellar brain inhibition by a decrease in activity of Purkinje cells <sup>34</sup>. Anodal cb-tDCS might positively interfere with this process when applied early <sup>34,35</sup>.

“POstural feedback ThErapy combined with Non-invasive TranscranIAL direct current stimulation in patients with stroke” (acronym POTENTIAL) aims to establish whether virtual reality based postural feedback training (VR-PFT) combined with anodal cb-tDCS is more effective than VR-PFT with sham cb-tDCS in improving standing balance, starting within 5 weeks post-stroke. Clinical measurements are needed to establish the clinical relevance of cb-tDCS, while posturographical and neurophysiological measurements are required to gain understanding into underlying mechanisms of standing balance performance and recovery post-stroke <sup>36</sup>. We hypothesise that: patients receiving VR-PFT+cb-tDCS will show a clinically meaningful improvement of 6 points or more on the Berg balance scale (BBS) at fifteen weeks post-stroke when compared to patients receiving VR-PFT+sham. A significantly larger decrease over time in centre of pressure (CoP) parameters is expected after VR-PFT+cb-tDCS as compared to VR-PFT+sham. We also hypothesise that these posturographical improvements will be accompanied by neurophysiological changes evident in normalisation in EEG-based theta and alpha power spectral density and cortical asymmetries between hemispheres.

## METHODS

### Study design

POTENTIAL is a double-blind randomised controlled trial, with fifteen intervention sessions of one hour during 3 weeks and a follow-up period until fifteen weeks post-stroke. Forty-six patients with a first-ever ischemic stroke will be enrolled within 5 weeks post-stroke. The study has been approved by the local medical ethical committee (NL52021.029.15), is registered in the Dutch trial register (NTR5261) and designed according to the criteria of the CONSORT 2010 statement <sup>37</sup>. A flowchart of the study procedures can be found in Figure 7.1.

### Patient population

Inclusion criteria are displayed in Figure 7.1.

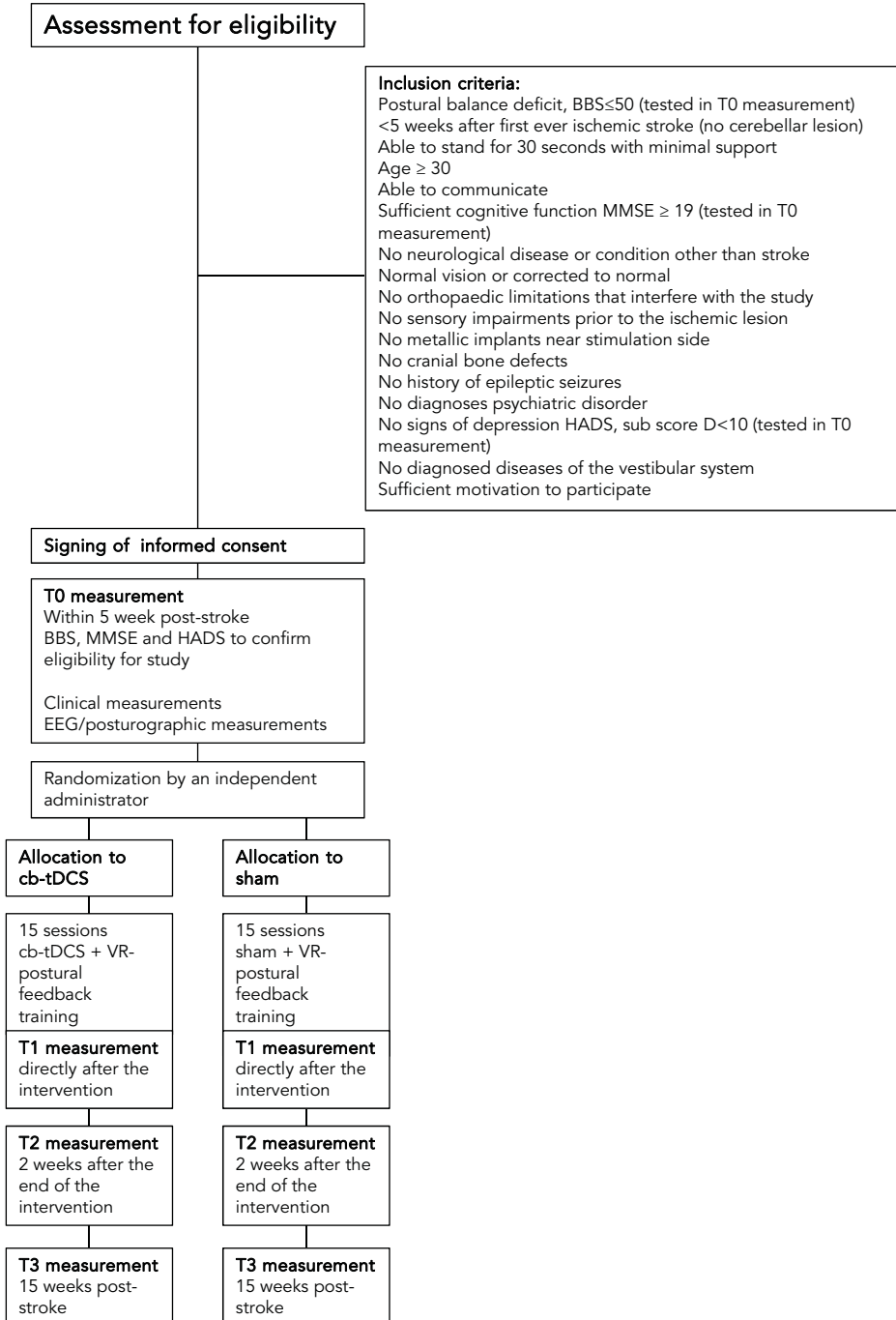


Figure 7.1 | Flowchart of the patient inclusion and study procedures.

Abbreviations: Berg balance scale (BBS), mini mental state examination (MMSE), hospital anxiety and depression scale (HADS).



## Randomisation and blinding procedure

Patient, assessors and therapists will be blinded to treatment allocation. Block randomisation per participating centre with blocks of 6 (last block of four) will be used. Concealed allocation will be effectuated with an online randomisation tool (Julius center, Utrecht, the Netherlands) performed by an independent administrator who will convey the randomisation into the tDCS software per patient. The group allocation is secured by a code only known to the independent administrator.

## Intervention

Training and measurements will take place at the rehabilitation facility where patients reside or receive outpatient therapy. Fifteen VR-PFT sessions, applied 5 days per week for 1 hour will be started within 5 weeks post-stroke, in addition to usual care. Subjects will be randomised by an independent administrator, into either VR-postural feedback training plus active cb-tDCS (N=23) or VR-postural feedback training plus sham (N=23). VR-PFT will be given by trained physical therapists on a balance workstation (Motek, Amsterdam, the Netherlands). The balance workstation consists of a customised software setup with a computer and 42 inch flat screen TV on a frame. VR software applications will be implemented in which visual feedback is given regarding centre of gravity or trunk movements during several tasks requiring active control of posture and balance in a virtual environment (D-flow, Motek, Amsterdam, the Netherlands), see Supplementary Table S7.1.

## Cb-tDCS application

tDCS will be applied starting 5 min before and during the first 20 min of each training session. The stimulation will be delivered by a portable stimulator (Starstim®, Neuroelectronics, Barcelona, Spain) through a pair of 3.14 cm<sup>2</sup> electrodes filled with a conducting gel, see Figure 7.2.

The anodal electrode will be placed 3 cm lateral of the inion towards the affected leg side, the cathodal electrode over the buccinator muscle. A 1.5 mA constant current will be applied in the cb-tDCS-group for 25 min with a ramp up and down phase of 30 s. The sham-group will receive a 0.5 mA ramp up of 30 s followed by a ramp down of 30 s, 24 min of 0 mA current ending with a 0.5 mA ramp up of 30 s and a ramp down of 30 s. Sham stimulation is a common procedure in tDCS research as an effective and reliable blinding method<sup>38</sup>.



Figure 7.2 | Head cap with portable wireless tDCS stimulator.

### Measurement outline

Assessments will be carried out prior to treatment allocation (baseline assessment, T0) as well directly after the intervention (T1), repeated 2 weeks after the end of the intervention (T2) and at 15 weeks post-stroke (T3). The clinical measures are performed by the researchers according to recommended guidelines<sup>39</sup> covering the three domains of the international classification of functioning, disability and health<sup>40</sup>.

### Primary outcome measure

The main outcome parameter is the Berg balance scale, which assesses balance performance and consists of fourteen test items in which the patient is asked to maintain a number of standing positions and to perform a number of balance tasks of increasing

difficulty. The test is reliable and valid in stroke patients<sup>41</sup>. A 6 points change is considered a clinical relevant difference<sup>42</sup>.

### Secondary outcome measures

The secondary measures performed are: Fugl-Meyer motor assessment of the lower extremity<sup>43</sup>, Motricity index arm and leg<sup>44</sup>, Erasmus modifications to the Nottingham sensory assessment of both legs<sup>45</sup>, fall history, 10-m walk test<sup>46</sup>, falls efficacy scale<sup>47</sup>, Nottingham extended activities of daily living<sup>48</sup>, stroke impact scale version 3.0<sup>49</sup>.

Patient descriptors: age, date of stroke, affected side, Bamford classification, comorbidities, handedness and smoking habits will be recorded at T0.

### Posturographic assessment

Ground reaction forces will be measured to assess standing balance performance, see Figure 7.3 for the complete setup. A monitor providing VF is positioned at eye-height in front of two force platforms, one foot positioned on each plate (Motek, Amsterdam, the Netherlands).

The following conditions will be tested:

1. Sit eyes open: to obtain the resting state activity of the brain, four times 60 s of EEG will be recorded while the patient is seated and is asked to look at a dot in front of him/her. 2 min will be recorded at the beginning and 2 min at the end of the session.
2. Quiet stance eyes-open/ eyes-closed: the patient will be asked to stand on the force platforms. Five trials with eyes open and 5 trials with eyes closed will be recorded for 60 s.
3. Tandem stance: the patient is asked, to hold the most difficult position that is feasible to perform for minimally 30 s. Five trials will be performed.
4. Anterior-posterior and medio-lateral limits of stability: the patient is asked to shift his/ hers CoP forward-backward and sideways as much as possible shown by a moving dot on a video screen while maintaining the same foot position.

### Posturographic outcome measures

CoP time series will be used to calculate qualitative measures of standing balance performance. For the quiet stance conditions we will determine: mean amplitude, amplitude variability, range, velocity, variability of the velocity and a composite-score of the above mentioned parameters representing standing balance performance<sup>33,50</sup>. The anterior-



Figure 7.3 | Balance workstation with double force platforms, visual feedback and concurrent measured EEG.

posterior and medio-lateral limits will be used to determine the area of the patients limits of stability.

### Neurophysiological assessment and outcome measures

During the quiet stance conditions and sitting task, 32-channel EEG will be recorded. Electrodes will be placed onto the skull using a head cap according to the international 10–20 system (TMSI International, Enschede, the Netherlands). Line noise will be reduced via bandpass filters and artefacts will be removed by an independent component analysis

approach<sup>51</sup>. The Fieldtrip toolbox for MEG and EEG analysis will serve to estimate power spectral densities<sup>52</sup>. Spectral power in the theta band (4–8 Hz), alpha band (8–12.5 Hz) and the beta band (15–30Hz) will be calculated. Asymmetry between hemispheres will be quantified with the brain symmetry index<sup>19,53</sup>.

### Sample size calculation

Sample size of this phase II study was calculated using a two-sided alpha of 0.2 with a power of 80% to correctly identify a potentially beneficial intervention<sup>54,55</sup>.

Previous studies among (sub)acute ischemic stroke patients, have reported BBS values with a mean of (M): 10, standard deviation (SD): 10<sup>56</sup> and median (med): 12, interquartile range (iqr): 2–22<sup>57</sup>. VR-PFT+cb-tDCS provides benefit over VR-PFT if the improvement on the BBS over time is 6 points larger at 15 weeks post-stroke<sup>42</sup>. In order to find a 6 points difference in improvement, with a SD of 11, 19 patients per group are needed. Using a 15% inflation to allow for non-parametric testing, and allow for a 10% loss to follow-up, we will need to enrol 23 patients per group.

### Statistical analysis

The BBS as the main outcome parameter of this study will be analysed with a Mann-Whitney-U test to establish a possible difference between the cb-tDCS and the sham group. The null hypothesis will be rejected if the cb-tDCS group shows a larger increase in BBS score between T0 and T3, as compared to the sham group with a probability value lower than 0.05. The difference between the groups needs to be 6 points or larger to be clinically meaningful.

Secondary outcome measures will be analysed using a mixed-model approach to establish statistical differences over time and between stimulation groups. This models will include factor time (T0, T1, T2, T3) and stimulation group (cb-tDCS versus sham) for which T0 and sham will be used as contrast. The distribution of the data or residuals of the models will be tested for normality using the Shapiro-Wilk test and by visual inspection of the histogram when appropriate. When normality of the residual is not met or in case of ordinal data and transformation to meet these criteria do not apply, a nonparametric equivalent will be used. The null hypothesis will be rejected if the corresponding probability value in the cb-tDCS group in measurement T3 is lower than 0.05 for the BBS as the main outcome parameter of this study.

## Study organisation and funding

Patients will be included in Reade rehabilitation center Amsterdam and Vogellanden rehabilitation center Zwolle, the Netherlands. Research coordination and analyses will be conducted at Amsterdam UMC. The study is funded by the Dutch brain foundation, the Netherlands.

## DISCUSSION

This trial will contribute to further understanding of underlying post-stroke recovery processes and whether these can be influenced by tDCS. Thereby it will add to a current lack of translational models of preclinical to human studies which are needed for instance to explain the large individual variability previously observed in tDCS studies<sup>33,58,59</sup>. As has been recommended by the series of rehabilitation roundtable papers of the leading experts in the field, we will combine clinical, posturographical and neurophysiological measurements and conduct the follow-up measurement at a fixed time point to enhance understanding of post-stroke recovery<sup>21,22,60-63</sup>.

## Summary and conclusions

This proof of concept double blind, sham controlled trial will show whether VR-PFT combined with anodal cerebellar tDCS is more effective than VR-PFT with sham in improving standing balance, measured with the BBS, started within the critical time window for homeostatic neuroplasticity within 5 weeks post-stroke.

## Funding

The study is funded by a fellowship of the Dutch brain foundation (Hersenstichting), the Netherlands, awarded to dr. E.E.H. van Wegen. Grant nr: F2013(1)-41.

## REFERENCES





- 1 Bohannon RW, Leary KM. Standing balance and function over the course of acute rehabilitation. *Arch Phys Med Rehabil* 1995; **76**: 994–996.
- 2 Desrosiers J, Noreau L, Rochette A, Bravo G, Boutin C. Predictors of handicap situations following post-stroke rehabilitation. *Disabil Rehabil* 2002; **24**: 774–785.
- 3 Kwakkel G, Kollen B, Twisk J. Impact of time on improvement of outcome after stroke. *Stroke* 2006; **37**: 2348–2353.
- 4 Biernaskie J, Chernenko G, Corbett D. Efficacy of rehabilitative experience declines with time after focal ischemic brain injury. *J Neurosci* 2004; **24**: 1245–1254.
- 5 Murphy TH, Corbett D. Plasticity during stroke recovery: from synapse to behaviour. *Nat Rev Neurosci* 2009; **10**: 861–872.
- 6 Reis J, Fritsch B. Modulation of motor performance and motor learning by transcranial direct current stimulation. *Curr Opin Neurol* 2011; **24**: 590–596.
- 7 Galea JM, Jayaram G, Ajagbe L, Celnik P. Modulation of cerebellar excitability by polarity-specific noninvasive direct current stimulation. *J Neurosci* 2009; **29**: 9115–9122.
- 8 Galea JM, Vazquez A, Pasricha N, Orban De Xivry JJ, Celnik P. Dissociating the roles of the cerebellum and motor cortex during adaptive learning: The motor cortex retains what the cerebellum learns. *Cereb Cortex* 2011; **21**: 1761–1770.
- 9 Saeys W, Vereeck L, Lafosse C, Truijien S, Wuyts FL, Van De Heyning P. Transcranial direct current stimulation in the recovery of postural control after stroke: a pilot study. *Disabil Rehabil* 2015; **37**: 1857–1863.
- 10 Hunter MA, Coffman BA, Trumbo MC, Clark VP. Tracking the neuroplastic changes associated with transcranial direct current stimulation: a push for multimodal imaging. *Front Hum Neurosci* 2013; **7**: 495.
- 11 Hülsdünker T, Mierau A, Neeb C, Kleinöder H, Strüder HK. Cortical processes associated with continuous balance control as revealed by EEG spectral power. *Neurosci Lett* 2015; **592**: 1–5.
- 12 Del Percio C, Babiloni C, Marzano N, Iacoboni M, Infarinato F, Vecchio F et al. 'Neural efficiency' of athletes' brain for upright standing: A high-resolution EEG study. *Brain Res Bull* 2009; **79**: 193–200.
- 13 Zhuang P, Toro C, Grafman J, Manganotti P, Leocani L, Hallett M. Event-related desynchronization (ERD) in the alpha frequency during development of implicit and explicit learning. *Electroencephalogr Clin Neurophysiol* 1997; **102**: 374–381.
- 14 Rabiller G, He JW, Nishijima Y, Wong A, Liu J. Perturbation of brain oscillations after ischemic stroke: A potential biomarker for post-stroke function and therapy. *Int J Mol Sci* 2015; **16**: 25605–25640.
- 15 Cavanagh JF, Frank MJ. Frontal theta as a mechanism for cognitive control. *Trends Cogn Sci* 2014; **18**: 414–421.
- 16 Finnigan SP, Walsh M, Rose SE, Chalk JB. Quantitative EEG indices of sub-acute ischaemic stroke correlate with clinical outcomes. *Clin Neurophysiol* 2007; **118**: 2525–2532.
- 17 Burghaus L, Hilker R, Dohmen C, Bosche B, Winhuisen L, Galldiks N et al. Early electroencephalography in acute ischemic stroke: Prediction of a malignant course? *Clin Neurol Neurosurg* 2007; **109**: 45–49.
- 18 Cuspidina E, Machado C, Galán L, Aubert E, Alvarez M a, Llopis F et al. QEEG prognostic value in acute stroke. *Clin EEG Neurosci* 2007; **38**: 155–160.
- 19 Sheorajpanday RVA, Nagels G, Weeren AJTM, van Putten MJAM, De Deyn PP. Quantitative EEG in ischemic stroke: Correlation with functional status after 6 months. *Clin Neurophysiol* 2011; **122**: 874–883.
- 20 Van Putten MJAM, Tavy DLJ. Continuous quantitative EEG monitoring in hemispheric stroke patients using the brain symmetry index. *Stroke* 2004; **35**: 2489–2492.
- 21 Bernhardt J, Kwakkel G, Lannin NA, Borschmann K, English C, Ali M et al. Consensus Statements from the Stroke Recovery and Rehabilitation Roundtable Standardized Measurement of Sensorimotor Recovery in Stroke Trials: Consensus-Based Core Recommendations from the Stroke Recovery and Rehabilitation Roundtable. *J Stroke* 2017; **31**: 784–792.







- 22 Boyd LA, Hayward KS, Ward NS, Stinear CM, Rosso C, Fisher RJ et al. Biomarkers of Stroke Recovery: Consensus-Based Core Recommendations from the Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2017; **12**: 480–493.
- 23 Ward NS. Does neuroimaging help to deliver better recovery of movement after stroke? *Curr Opin Neurol* 2015; **28**: 323–9.
- 24 Pollok B, Butz M, Gross J, Südmeyer M, Timmermann L, Schnitzler A. Coupling between cerebellar hemispheres: Behavioural, anatomic, and functional data. *Cerebellum* 2006; **5**: 212–219.
- 25 Ito M, Yamaguchi K, Nagao S, Yamazaki T. Long-Term Depression as a Model of Cerebellar Plasticity. *Prog Brain Res* 2014; **210**: 1–30.
- 26 Carey MR. Synaptic mechanisms of sensorimotor learning in the cerebellum. *Curr Opin Neurobiol* 2011; **21**: 609–615.
- 27 Cheron G, Dan B, Marquez-Ruiz J. Translational approach to behavioral learning: lessons from cerebellar plasticity. *Neural Plast* 2013; **2013**: e853654.
- 28 de Rooij IJM, van de Port IGL, Meijer J-WG. Effect of Virtual Reality Training on Balance and Gait Ability in Patients With Stroke: Systematic Review and Meta-Analysis. *Phys Ther* 2016; **96**: 1905–1918.
- 29 Iruthayarajah J, McIntyre A, Cotoi A, Macaluso S, Teasell R. The use of virtual reality for balance among individuals with chronic stroke: a systematic review and meta-analysis. *Top Stroke Rehabil* 2017; **24**: 68–79.
- 30 Cano Porras D, Siemonsma P, Inzelberg R, Zeilig G, Plotnik M. Advantages of virtual reality in the rehabilitation of balance and gait: Systematic review. *Neurology* 2018; **90**: 1017–1025.
- 31 Caligiore D, Pezzulo G, Baldassarre G, Bostan AC, Strick PL, Doya K et al. Consensus Paper: Towards a Systems-Level View of Cerebellar Function: the Interplay Between Cerebellum, Basal Ganglia, and Cortex. *Cerebellum* 2017; **16**: 203–229.
- 32 Rampersad SM, Janssen AM, Lucka F, Aydin U, Lanfer B, Lew S et al. Simulating transcranial direct current stimulation with a detailed anisotropic human head model. *IEEE Trans Neural Syst Rehabil Eng* 2014; **22**: 441–452.
- 33 Zandvliet SB, Meskers CGM, Kwakkel G, van Wegen EEH. Short-Term Effects of Cerebellar tDCS on Standing Balance Performance in Patients with Chronic Stroke and Healthy Age-Matched Elderly. *Cerebellum* 2018; **17**: 575–589.
- 34 Gold L, Lauritzen M. Neuronal deactivation explains decreased cerebellar blood flow in response to focal cerebral ischemia or suppressed neocortical function. *Proc Natl Acad Sci* 2002; **99**: 7699–7704.
- 35 Sobesky J, Thiel A, Ghaemi M, Hilker RH, Rudolf J, Jacobs AH et al. Crossed cerebellar diaschisis in acute human stroke: a PET study of serial changes and response to supratentorial reperfusion. *J Cereb Blood Flow Metab* 2005; **25**: 1685–1691.
- 36 Kwakkel G, Lannin NA, Borschmann K, English C, Ali M, Churilov L et al. Standardized measurement of sensorimotor recovery in stroke trials: Consensus-based core recommendations from the Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2017; **12**: 451–461.
- 37 Schulz KF, Altman DG, Moher D. CONSORT 2010 Statement: updated guidelines for reporting parallel group randomised trials. *BMC Med* 2010; **24**: 18.
- 38 Garnett EO, den Ouden D-B. Validating a Sham Condition for Use in High Definition Transcranial Direct Current Stimulation. *Brain Stimul* 2015; **8**: 551–554.
- 39 Veerbeek J, Van Wegen E, Van Peppen R, Van Der Wees PJ, Hendriks E, Rietberg M et al. What is the evidence for physical therapy poststroke? A systematic review and meta-analysis. *PLoS One* 2014; **9**: e87987.
- 40 World Health organization. International Classification of Functioning, Disability and Health. Geneva, 2001.
- 41 Blum L, Korner-Bitensky N. Usefulness of the Berg Balance Scale in stroke rehabilitation: a systematic review. *Phys Ther* 2008; **88**: 559–566.
- 42 Stevenson TJ. Detecting change in patients with stroke using the Berg Balance Scale. *Aust J Physiother* 2001; **47**: 29–38.
- 43 Fugl-Meyer AR, Jääskö L, Leyman I, Olsson S, Steglind S. The post-stroke hemiplegic patient. 1. a method for evaluation of physical performance. *Scand J Rehabil Med* 1975; **7**: 13–31.

- 44 Collin C, Wade D. Assessing motor impairment after stroke: a pilot reliability study. *J Neurol Neurosurg Psychiatry* 1990; **53**: 576–579.
- 45 Stolk-Hornsveld F, Crow JL, Hendriks EP, Van Der Baan R, Harmeling-van Der Wel BC. The Erasmus MC modifications to the (revised) Nottingham Sensory Assessment: a reliable somato-sensory assessment measure for patients with intracranial disorders. *Clin Rehabil* 2006; **20**: 160–172.
- 46 Collen FM, Wade DT, Bradshaw CM. Mobility after stroke: reliability of measures of impairment and disability. *Int Disabil Stud* 1990; **12**: 6–9.
- 47 Tinetti ME, Richman D, Powell L. Falls efficacy as a measure of fear of falling. *J Gerontol* 1990; **45**: 239–243.
- 48 Nouri F, Lincoln N. An extended activities of daily living scale for stroke patients. *Clin Rehabil* 1987; **1**: 301–305.
- 49 Duncan PW, Lai SM, Bode RK, Perera S, DeRosa J. Stroke Impact Scale-16: A brief assessment of physical function. *Neurology* 2003; **60**: 291–296.
- 50 Pasma JH, Bijlsma AY, Van Der Bij MDW, Arendzen JH, Meskers CGM, Maier AB. Age-related differences in quality of standing balance using a composite score. *Gerontology* 2014; **60**: 306–314.
- 51 Jung TP, Makeig S, Westerfield M, Townsend J, Courchesne E, Sejnowski TJ. Removal of eye activity artifacts from visual event-related potentials in normal and clinical subjects. *Clin Neurophysiol* 2000; **111**: 1745–1758.
- 52 Oostenveld R, Fries P, Maris E, Schoffelen J-M. FieldTrip: Open source software for advanced analysis of MEG, EEG, and invasive electrophysiological data. *Comput Intell Neurosci* 2011; **2011**: e156869.
- 53 Finnigan S, van Putten MJAM. EEG in ischaemic stroke: Quantitative EEG can uniquely inform (sub-)acute prognoses and clinical management. *Clin Neurophysiol* 2013; **124**: 10–19.
- 54 Schoenfeld D. Statistical considerations for pilot studies. *Int J Radiat Oncol Biol Phys* 1980; **6**: 371–374.
- 55 Stallard N. Optimal sample sizes for phase II clinical trials and pilot studies. *Stat Med* 2012; **31**: 1031–1042.
- 56 Ng MFW, Tong RKY, Li LSW. A pilot study of randomized clinical controlled trial of gait training in subacute stroke patients with partial body-weight support electromechanical gait trainer and functional electrical stimulation: Six-month follow-up. *Stroke* 2008; **39**: 154–160.
- 57 Allison R, Dennett R. Pilot randomized controlled trial to assess the impact of additional supported standing practice on functional ability post stroke. *Clin Rehabil* 2007; **21**: 614–619.
- 58 Ward NS. Restoring brain function after stroke - bridging the gap between animals and humans. *Nat Rev Neurol* 2017; **13**: 244–255.
- 59 Wiethoff S, Hamada M, Rothwell JC. Variability in response to transcranial direct current stimulation of the motor cortex. *Brain Stimul* 2014; **7**: 468–475.
- 60 Bernhardt J, Borschmann K, Boyd L, Thomas Carmichael S, Corbett D, Cramer SC et al. Moving rehabilitation research forward: Developing consensus statements for rehabilitation and recovery research. *Int J Stroke* 2016; **11**: 454–458.
- 61 Bernhardt J, Hayward KS, Kwakkel G, Ward NS, Wolf SL, Borschmann K et al. Agreed definitions and a shared vision for new standards in stroke recovery research: The Stroke Recovery and Rehabilitation Roundtable taskforce. *Int J Stroke* 2017; **12**: 444–450.
- 62 Corbett D, Carmichael ST, Murphy TH, Jones TA, Schwab ME, Jolkkonen J et al. Enhancing the alignment of the preclinical and clinical stroke recovery research pipeline: Consensus-based core recommendations from the Stroke Recovery and Rehabilitation Roundtable translational working group. *Int J Stroke* 2017; **12**: 462–471.
- 63 Walker MF, Hoffmann TC, Brady MC, Dean CM, Eng JJ, Farrin AJ et al. Improving the development, monitoring and reporting of stroke rehabilitation research: Consensus-based core recommendations from the Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2017; **12**: 472–479.


Supplemental Table S6.1 | Overview of the applications used for the Postural Feedback Training in this study

	Title	Maze 1 & 2
	Goal	Eliciting mediolateral (ML) and anterior-posterior (AP) movements of the patients centre of pressure.
	Outcome	Time and number of collisions with the wall.
	Instructions to patient	Move the red dot through the maze as fast as possible by shifting your weight forward/backwards/left/right. Try not to collide with any wall.
	Adjustable parameters	Maximal speed, sensitivity in ML and AP direction.
	Title	City Ride
	Goal	Eliciting ML and AP movements of the patients centre of pressure.
	Outcome	Time and number of collisions with the wall.
	Instructions to patient	You are driving a car through the city, try go get as far as possible and avoid oncoming traffic. Move the car left and right by shifting your weight accordingly and control the speed by shifting your weight forward and backward.
	Adjustable parameters	Lock AP movements (fixed speed), time, max speed, sensitivity ML and AP direction, percentage of oncoming traffic per side.
	Title	Reach the skies
	Goal	Eliciting ML and AP movements of the patients centre of pressure.
	Outcome	Number of blocks you catch in the set time.
	Instructions to patient	On top of the totem pole you see a grey figure, that is you. You can move left and right by shifting your weight accordingly. Try to align the red and grey figure as accurately as possible, this way you will be able to catch the falling blocks, heightening the totem pole.
	Adjustable parameters	Time, speed of blocks, allowed error.
	Title	Hole in the bridge
	Goal	Eliciting ML movements of the patients centre of pressure.
	Outcome	Number of rabbits that crossed the bridge.
	Instructions to patient	Try to save as many rabbits as possible by closing the gaps in the bridges with the boat. You can move the boat left and right by shifting your weight accordingly.
	Adjustable parameters	Time, rabbit speed, sensitivity, lock boat from moving more lateral that the two outer bridges.

Supplemental Table S6.1 | *Continued*

Title		Walk the line
	Goal	Eliciting ML and AP movements of the patients centre of pressure.
	Outcome	Time and percentage of precision following the white line
	Instructions to patient	Move the blue dot by shifting your weight. Push the orange dot with the blue dot. Try to follow the white line as accurately as possible for a high score.
	Adjustable parameters	Size orange dot, ML and AP sensitivity. Shapes: circle, triangle, heart, star, square, flower, elephant.
Title		Hit the mole
	Goal	Eliciting ML and AP movements of the patients centre of pressure.
	Outcome	% of moles hit
	Instructions to patient	Moles will pop up from the holes in the ground, try to hit them with the hammer by putting weight with your leg at the appointed space on the force platform.
	Adjustable parameters	Speed and time.
Title		The boat
	Goal	Eliciting ML and AP movements of the patients centre of mass.
	Outcome	Time
	Instructions to patient	Try to steer the boat pass the buoys as fast as possible. Move the boat left and right by shifting your weight accordingly. Leaning forward means speeding up and leaning backward is slowing down.
	Adjustable parameters	Speed and sensitivity.
Title		Paper flight
	Goal	Eliciting ML and AP movements of the patients centre of mass.
	Outcome	Passed white rings
	Instructions to patient	Try to fly the plane through the tunnel made of rings. Move the plane by moving your trunk. Leaning forward means diving and leaning backward is an upward movement. For each passed white ring a point is given.
	Adjustable parameters	Time, speed, sensitivity.

Supplemental Table S6.1 | *Continued*

	Title	Sit to stand
	<p>Goal</p> <p>Outcome</p> <p>Instructions to patient</p> <p>Adjustable parameters</p>	<p>Practicing standing up.</p> <p>Alien is taken by the spaceship.</p> <p>Try to stand up independently. If you move the trunk forward, the alien will walk across the planet. If you stand upright, the alien will be taken by the space ship. Make sure to straighten and put weight on the effected leg.</p> <p>Trunk angle.</p>





# Chapter 8

General discussion





The processes by which the central nerve system can learn, restore, and adapt to lost sensorimotor function after stroke are complex. Translational research is needed to improve our understanding of patients' time course of neurological and behavioural recovery early post-stroke. We aimed to strengthen the phenomenological model presented in Figure 1.1 using different approaches in addition to clinical assessments. System identification techniques, regression modelling, neurophysiological measures and non-invasive stimulation of the brain were used in this thesis to investigate the relation between underlying neuronal mechanisms, panel 1.1B, to processes that contribute to sensorimotor recovery after stroke, panel 1.1A. In this chapter, the main findings are discussed. Recommendations for future translational research to improve our understanding of individualized post-stroke recovery processes are given.

## PART 1: Spontaneous neurobiological recovery after stroke

### The influence of somatosensory impairment on motor recovery

The first part of this thesis, Figure 1.1 blue panel, focusses on non-learning dependent mechanisms and spontaneous neurobiological recovery after stroke.

In chapter 2, the influence of somatosensory impairments, on behavioural restitution of upper limb motor function was investigated. A number of clinical observational studies suggest that a severe somatosensory impairment may hamper the recovery of motor function<sup>1,2</sup>. In chapter 2 we therefore investigated if the relationship between motor and somatosensory recovery could reflect a parallel recovery in both modalities in the first 6 months post-stroke and if intactness of somatosensory function is a pre-requisite for upper limb motor recovery early post-stroke. We hypothesized that improvements in both motor and somatosensory impairments were largely driven by common processes of spontaneous neurobiological recovery. In line with previous prospective cohort studies<sup>3-5</sup>, we observed a somatosensory impairment in about 50% of the 215 measured patients with a first-ever ischemic stroke. When analysing the population of 94 patients with somatosensory impairments, the significant association between motor and somatosensory recovery disappeared when correcting for the progress of time. This finding suggests that spontaneous neurobiological recovery is an important factor that drives both motor and somatosensory recovery of the upper limb early post-stroke. Since the association between motor and somatosensory recovery for patients with severe somatosensory impairment at baseline did not significantly differ from the whole group, we concluded that somatosensory impairment at baseline does not compromise motor recovery, per se. In contrast, we found significant differences in the association between motor and somatosensory recovery when separately

looking at patients who show motor recovery (recoverers) and patients who show little to no motor recovery (non-recoverers). Motor recovery was found to be positively associated with somatosensory recovery, independent of progress of time, in patients who had a mild motor impairment or showed an improvement of 6 points or more on the Fugl-Meyer motor assessment of the paretic upper extremity (FM-UE) at 26 weeks post-stroke (recoverers). In addition to mechanisms that drive spontaneous neurobiological recovery in both modalities, recovery of somatosensory impairment seems to be conditional for full behavioural restitution of motor recovery. Associations between modalities can be concealed if different recovery patterns are not taken into account as it is the case in a normal association model. By applying a hybrid association model, we distinguished within- and between-subject effects, showing that outcomes are robust for the influence of arbitrary cut-off scores <sup>6</sup>. In particular, when the cut-offs to identify subgroups are not clearly defined, a hybrid association model can be of help to disentangle the complex interplay of within- and between-subject effects during the time course of spontaneous neurobiological recovery early post-stroke.

### **Predicting spontaneous neurobiological recovery**

The proportional recovery model provides a broad distinction between patients who show recovery of 64% <sup>7,8</sup> up to 97% <sup>9</sup>, of their initial impairment depending on the modality, so-called 'fitters' or 'recoverers', and patients who show very poor outcome of function in the chronic phase and little to no improvement over time, so-called 'non-fitters' or 'non-recoverers' <sup>7,9-14</sup>. The model seems to overestimate the predictability of recovery since baseline and recovery (i.e. the difference between baseline and outcome at 6 months) are more strongly correlated than baseline and outcome at 6 months <sup>15</sup>. However, it is not likely that this overestimation influences the distinction into patients who show (recoverers) and patients who do not show any or very little recovery (non-recoverers) <sup>16</sup>. This classification of recoverers and non-recoverers can give support to understand the mechanisms that drive recovery and help the development of accurate prediction models for outcome. Importantly, very different research and treatment approaches are needed for those patients who do show some spontaneous neurological recovery early post-stroke when compared to those in which spontaneous neurobiological recovery is completely absent. Innovative prognostic mixture models may be the next steps to develop early individual clinical decision making at stroke units <sup>17</sup>. In addition, these mixture models may further identify subgroups of proportional recovery and capture the observed variability in time windows of spontaneous recovery <sup>17</sup>, see Figure 8.1 for recommendations. A patient's ability to perceive and modulate, for example due to somatosensory impairment, is an important factor to consider in these models, see Figure 1.1 in the general introduction.

In clinical care, age is often used in triage of patients into rehabilitation or nursing facilities and is mentioned as a possible variable in prediction models. The Copenhagen study for example, found that patients had an average of a 3 points lower score on the Barthel Index at hospital discharge for every 10 years of higher age<sup>18</sup>. However, this finding does not suggest that age is an important factor for the amount of spontaneous neurobiological recovery<sup>18</sup>. In the same vein, our results also indicate that higher age on itself is not a key factor for predicting the amount of sensorimotor improvement in patients with a first-ever ischemic stroke. Above findings rather suggests that age is associated with pre-stroke comorbidities measured at baseline in our association model. Factors that show high collinearity with higher age, such as pre-existing comorbidities as diabetes and mobility problems<sup>18,19</sup> are suggested to be more important for understanding recovery than the influence of age on neurobiological recovery itself. In other words, the potential for neurobiological recovery in subjects with high age without comorbidities, might not be different when compared to the recovery of a younger person with a stroke.

### **Alternatives for clinical assessment of somatosensory impairments**

One of the reasons for the limited attention that somatosensory recovery has received in post-stroke research might be the absence of a golden standard assessment tool to measure somatosensory impairment and recovery. While the Erasmus modification of the Nottingham sensory assessment (EmNSA) has a good to excellent reliability<sup>20,21</sup>, the smallest detectable change or minimal clinically relevant difference of this scale, have not been determined. It can be hard to obtain accurate and valid EmNSA scores for patients with cognitive or attention impairments. Clinical practice would highly benefit from measures that can more objectively establish somatosensory function. Neurophysiological and imaging techniques such as DTI, Somatosensory Evoked Potentials as well as measuring cortical coherence by wrist position perturbations may be more objective method than clinical somatosensory scales. As shown in **chapter 4 and 5** of this thesis, position-cortical coherence (PCC), or somatosensory and median nerve stimulation to the affected arm<sup>22</sup>, as valid and precise ways to test the integrity of somatosensory pathways after stroke<sup>22-24</sup>. The prognostic value of these non-invasive techniques above clinical testing alone needs further investigation.

In addition, to understand mechanisms of recovery, longitudinal studies with fixed measurement points in time, may further elucidate the non-linear dynamics of motor and somatosensory recovery within the time window of spontaneous neurobiological recovery.

## Neurophysiological markers of cortical reorganisation

### *Resting-state spectral characteristics*

Several specific frequency ranges of neuronal oscillations measured with EEG and MEG have been linked to specific behaviour in humans and animals<sup>25</sup>. Increased power of slow oscillations are consistently found after stroke and correlates with stroke severity and a larger infarction volume<sup>26–33</sup>. In **chapter 3**, EEG was longitudinally measured in 41 patients during awake rest to evaluate spectral characteristics as a representation of cortical reorganisation and their development over time in relation to recovery of motor function and stroke severity. It was hypothesized that an increase in slow oscillations and a relative larger asymmetry in spectral characteristics between hemispheres in the first weeks after stroke were related to the stroke severity and motor function of the upper limb. We found increased spectral power in the low frequency delta band of the affected hemisphere, which is in agreement with literature<sup>34</sup>. The spectral power in the delta band was increased relative to alpha activity, i.e. delta-alpha ratio of the affected hemisphere ( $DAR_{AH}$ ), and relative to the unaffected hemisphere, i.e. directional BSI ( $BSI_{dir_{delta}}$ ), within 3 weeks post-stroke and gradually decreased over time.  $DAR_{AH}$  and  $BSI_{dir_{delta}}$  were associated with stroke severity, measured with the national institute of health stroke scale, within 3 weeks post-stroke. Neural tissue affected by the stroke is likely responsible for this decrease in frequency of neuronal oscillations<sup>34</sup>. With that, the amount of delta increase seems to reflect the severity of pathology in the brain.  $DAR_{AH}$  and  $BSI_{dir_{delta}}$  showed potential as markers in future prediction models, reflecting spontaneous neurobiological recovery over time within a person and can potentially distinguish between patients with different recovery patterns. However, resting-state spectral characteristics that quantify the strength of the neural oscillations seem to be a rather global measure of pathology in the brain. Our study suggests that in order to capture cortical reorganisation related to sensorimotor recovery, the motor system itself might need to be studied by using passive and active sensorimotor tasks. For patients with severe upper limb impairments with absence of voluntary motor control, such a task can obviously only be passive. Furthermore, we showed in **chapter 5** that closed-loop system identification techniques may help to investigate sensorimotor intactness.

### *Position-cortical coherence*

The cortical response to passive wrist joint movement can be measured consistently in healthy subjects, as well as in severely affected patients. Such a passive wrist joint movement specifically perturbs the proprioceptive system, which is highly involved in motor execution tasks. **Chapters 4 and 5** evaluated afferent sensory pathway integrity

and information processing after stroke with the parameter position-cortical coherence (PCC), representing the coherence between cortical activity, measured with EEG, and a mechanically evoked wrist position perturbation in the affected limb. The presence of PCC (%PCC) was hypothesized as a potential neurophysiological marker of somatosensory pathway integrity related to spontaneous neurobiological recovery.

In the cross-sectional study described in **chapter 4**, all 11 stroke patients showed significant contralateral PCC. Patients with poor motor function had a reduced contralateral PCC as compared to patients with good motor function in the affected wrist. In **chapter 5** we longitudinally evaluated PCC to capture its dynamics in the time window of spontaneous neurobiological recovery in 48 patients after stroke and its relation with somatosensory and motor recovery. %PCC increased from baseline to 12 weeks post-stroke, which was in accordance with the recovery seen on the EmNSA and FM-UE scales. A significant positive association was found between %PCC, mean amplitude in the affective hemisphere (Amp-A) and EmNSA of the hand and fingers. PCC is closely related to the motor system and shows a change as expected in the time window of spontaneous neurobiological recovery. PCC was higher at baseline in terms of percentage and amplitude in patients who showed some motor recovery at 26 weeks post-stroke as compared to patients with poor or no recovery. Yet the PCC measure did not solely seem to represent the information transfer across the afferent pathways. The used perturbation also seems to evoke a cortical response in all patients. We used a perturbation signal that contained frequencies between 5 and 29 Hz, which overlaps for a large part with the beta band (15–30 Hz). Neuronal oscillations, particular in the beta band, are sensitive to gamma-Aminobutyric acid (GABA)-ergic and glutamatergic processes which are enhanced in the time window of spontaneous neurobiological recovery and are important for neural plasticity and recovery after stroke<sup>25,35,36</sup>. Since a lower power in the beta frequencies correlates with a large lesion size in patients within one day after stroke<sup>37</sup>, a lower response to the applied perturbation signal in the beta band could reflect pathology in the brain. To investigate this interpretation of our results, a smaller range of perturbed frequencies could be investigated in the future, which should separate alpha and beta band frequencies from slower neural oscillations. Non-linear coherence measures could also provide insights into the highly non-linear responses of neural pathways to a wrist joint perturbation<sup>38,39</sup>. For PCC, and other markers based on the coherence of neural responses and oscillations, to have a role in prediction of function post-stroke, we need future studies to separate characteristics which give information on the initial severity of the stroke from the ones which provide information on subsequent recovery processes<sup>35</sup>.

### *Possible markers for somatosensory recovery*

Within this thesis, somatosensory recovery was mainly studied as a possible marker for motor recovery after stroke. However, there are only very limited candidate markers that give insight into the severity of the somatosensory impairment itself<sup>40-42</sup>. Boccuni et al. reported a greater lesion load in the corticospinal tract of patients with somatosensory impairments in terms of perception of stimuli at 4–7 days post-stroke, yet all of the 32 included patients showed full recovery of somatosensory perception at 26 weeks post-stroke<sup>42</sup>. This result indicates the important difference between markers of initial severity and markers of recovery function that are needed in the clinical practice<sup>35</sup>. In Figure 8.1, we give an overview of the mechanisms that are hypothesized to play a role in the time course of post-stroke recovery as well as the recommendations for further studies. PCC showed a longitudinal association with somatosensory recovery as measured with the EmNSA. Future studies are needed to investigate the predictive capacity of PCC as a marker of somatosensory recovery while disentangling the influence of somatosensory impairment on motor recovery that goes beyond common underlying mechanisms responsible for spontaneous neurobiological recovery in both domains.

## **PART 2: Modulation of sensorimotor recovery after stroke**

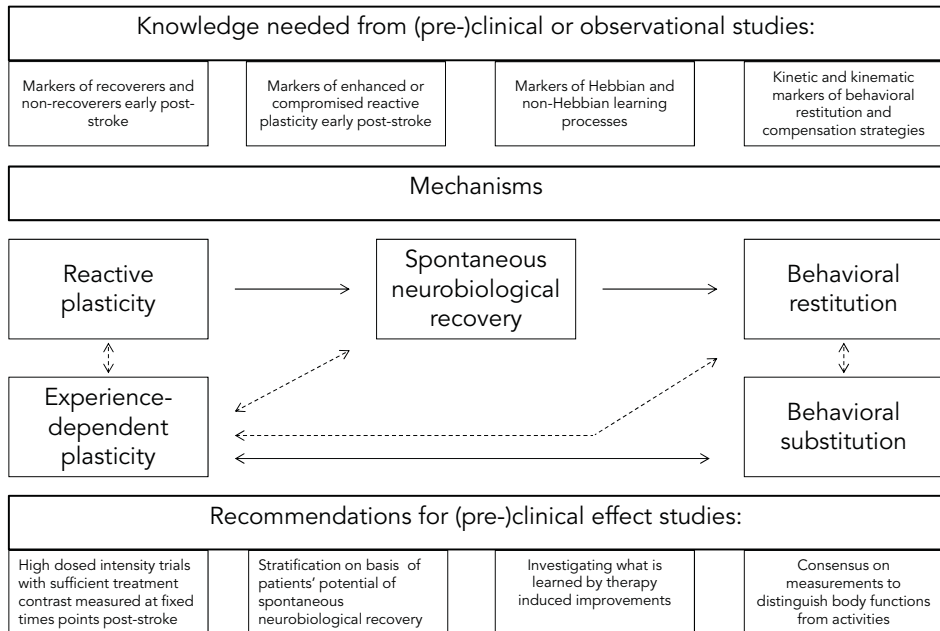
Next to understanding spontaneous neurobiological recovery after stroke, a key challenge is to optimize learning-dependent mechanisms and ultimately understand if reactive and experience-dependent plasticity can influence each other (see Figure 8.1, mechanisms). The second part of this thesis, Figure 8.1 green panel, focusses on learning-dependent mechanisms and the potential for modulation of sensorimotor recovery after stroke by brain stimulation (cerebellar transcranial direct current stimulation, cb-tDCS).

### **Interindividual differences in cerebellar tDCS response**

In **chapter 6** of this thesis, standing balance performance was investigated as a target for enhanced experience-dependent plasticity by cb-tDCS. A group of 15 patients with a chronic stroke received anodal cb-tDCS on the ipsilesional cerebellar hemisphere or contralateral cerebellar hemisphere as compared to sham, during a dynamic postural feedback-based tracking task. After receiving ipsilesional cb-tDCS, we found an instantaneous positive effect on a composite centre of pressure measure of stability of standing balance, but only in the most difficult tandem stance position.

Although changes in balance performance were observed at group level in **chapter 6**, high interindividual variability in response to cb-tDCS was found, which indicates that





**Figure 8.1** | Challenges and recommendations for post-stroke research.

Schematic overview of the mechanisms that are hypothesized to play a role in the time course of post-stroke recovery. The solid lines represent interactions between mechanisms that are assumed to take place, while the dashed lines represent possible interaction for which evidence in humans with a stroke is still lacking.

A key challenge is to disentangle how reactive and experience-dependent plasticity interact during recovery after stroke. To investigate the effect on spontaneous neurobiological recovery, markers of reactive and experience-dependent plasticity are essential, as well as reliable and responsive functional outcome measures for which prospective observational cohort studies are required.

(pre-)Clinical effect studies are required, to reveal if reactive and experience-dependent plasticity can influence each other and if this interaction can ultimately lead to enhanced spontaneous neurobiological recovery and therefore behavioural restitution. To understand if it is possible to intervene in these processes, recommendations for effect studies are given.

the exact mechanisms by which cb-tDCS enforces its effects are not clear. Intrinsic factors such as age, gender, and genetic differences, as well as extrinsic factors such as electrode placement, current intensity, stimulation duration<sup>43,44</sup>, the difficulty of the behavioural task<sup>45</sup> and smoking before a measurement<sup>46</sup>, are mentioned as possible factors that contribute to interindividual differences. Anatomical fibre orientation, connectivity between brain regions and the initial state of the neuronal population also have been reported to play a role in the variability of the response<sup>47-50</sup>. In order to fully take advantage of the potentially enhancing effects of tDCS on experience-dependent plasticity by LTP and LTD-like mechanisms, it is essential to have thorough evidence and understanding of the

mechanisms by which tDCS can enforce its effects<sup>51</sup> and from that point of view, explain and understand interindividual variability. Understanding which cells are polarized, and which compartments within these cells and their orientation relative to the current flow is essential to characterize tDCS effects<sup>52</sup>. Looking at the mechanisms of action of tDCS, it is evident that parameters of the stimulation protocol strongly determine the direction of synaptic plasticity and therefore the effects of the stimulation<sup>53</sup>. To tackle the issue of the large variability in responses, we need to formulate hypotheses on the intended neuronal changes and test these by combining non-invasive stimulation with neurophysiological measurements and imaging<sup>54,55</sup>.

In the cerebellum, a complex interplay of LTD and LTP of Purkinje cells is required for synaptic based forms of plasticity<sup>56-59</sup>. Parallel fibers and climbing fibers are suggested to have a mediating role in this process and give input to error signals in motor execution<sup>60-62</sup>. It has been suggested that anodal cb-tDCS enlarges the population of activated Purkinje cells, leading to a larger involvement of the cerebellum in the executed motor task<sup>63-65</sup>. More animal model studies are required to confirm the hypothesized mechanistic pathways of cb-tDCS<sup>66</sup>.

To achieve reproducible effects beyond neurophysiological excitability changes, thorough understanding of behavioural effects and learning mechanisms are also required<sup>67,68</sup>. Kinematic measures that can distinguish between behavioural restitution and substitution of function are needed to measure the subtle effects that can be expected from tDCS<sup>69,70</sup>.

### **Mechanisms of action of tDCS in patients with a stroke**

Designing an optimal protocol for non-invasive brain stimulation after stroke is difficult due to the unknown effects of the lesion on the current flow. A certain baseline degree of neuronal activity, which could be affected after stroke, is likely to be needed for synaptic plasticity since NMDA receptors need to be active<sup>71</sup>. The time window of heightened neural plasticity early after stroke<sup>72</sup> might at the other hand, give a unique chance to induce clinically relevant effects, which are unattained by therapy alone. In studies investigating the effect of cortical tDCS in patients after stroke, the hypothesized mechanisms of action have often been based on the interhemispheric competition model. The idea of this model is based on the mutual inhibiting effect of both hemispheres on each other, which has been disrupted due to the stroke<sup>73</sup>. In chronic stroke patients, this increased interhemispheric inhibition (IHI) of the non-affected on to the affected hemisphere prior to movement execution is correlated to poor motor recovery and might impair recovery<sup>74,75</sup>. Di Pino et al. suggested that IHI might not be a maladaptive mechanism of the brain after

stroke, yet rather a consequence of limited residual capacity in the affected hemisphere <sup>76</sup>. Recent studies showed that there is no evidence of IHI in the first 3 months post-stroke, by that disproving the interhemispheric competition model as being the cause of poor motor recovery after stroke and the hypothesized mechanisms of action behind many tDCS studies <sup>77,78</sup>. Whether the mechanisms of action of tDCS are stroke specific or represent similar effects as have been induced in the healthy brain can currently not yet be concluded. Longitudinal studies that combine behavioural with neurophysiological and imaging measures, starting in the acute phase after stroke, could give valuable insight into the cause and effect of motor recovery after stroke and the mechanisms by which tDCS enforces its effects.

Anodal cb-tDCS has been proposed as a mechanism that potentially can counteract the effects of crossed cerebellar diaschisis <sup>79,80</sup>, which induces a disbalance in cerebellar brain inhibition by a decrease in activity of Purkinje cells <sup>81</sup>. This specific hypothesis needs to be investigated by combining cb-tDCS with imaging methods to study diaschisis and network changes in the time window of spontaneous neurobiological recovery.

### **Modulation of sensorimotor recovery in the time window of spontaneous neurobiological recovery by cerebellar tDCS**

In the first 4–10 weeks post-stroke, neurotrophic factors create a critical time window for recovery <sup>72,82,83</sup>. If tDCS is able to accelerate experience-dependent plasticity, this effect has to be evident in this specific time window of enhanced reactive plasticity <sup>59,72</sup>.

The randomized double-blind and sham-controlled POTENTIAL trial, of which the protocol is described in **chapter 7** aims to determine if cb-tDCS applied in the early phase after stroke can enhance experience-dependent plasticity in the time window of spontaneous neurobiological recovery. The POTENTIAL trial is currently underway and designed to investigate a possible influence of experience-dependent plasticity on behavioural restitution of function. The effect of tDCS is studied with a combination of clinical, neurophysiological and posturographical measurements to learn more about the underlying mechanisms by which cb-tDCS enforces its effects and follows the recommendations for clinical effect studies (see Figure 8.1). Although this randomized controlled trial will not directly contribute to the neurobiological evidence, it can contribute to the translation of preclinical models into human studies, which is lacking at this moment <sup>84</sup>. Combining behavioural outcome measures with measures of diaschisis and functional network changes will enable us to relate hypothesized mechanisms of action of enhanced experience-dependent plasticity in the time window of spontaneous neurobiological

recovery. Before tDCS or other non-invasive brain stimulation could have a place in clinical care of patients with a stroke, the mechanisms including the dose-responds need to be better understood and effects need to be reproducible.

### **Transcranial alternating current stimulation; as an alternative for tDCS**

With tDCS, we can enhance or decrease the excitability of neurons in a certain area. However, the timing and synchronisation or desynchronization of neuronal populations are not directly influenced by tDCS. Synchronisation or desynchronization of neuronal activity, which can be measured as neuronal oscillations with different ranges of frequencies, has shown to play an important role in motor and cognitive functions<sup>35</sup>. If specific neuronal oscillations underlie the transfer of information in the brain, and transcranial alternating current stimulation (tACS) can entrain these intrinsic neuronal oscillations, it could have a much larger impact than what is currently achieved by tDCS<sup>85</sup>. Next to that, tACS applied to the cerebellum could be particularly interesting due to its distinct function in motor adaptation and timing. tACS might give us the opportunity to causally link brain oscillations of a specific frequency range to specific processes<sup>86</sup>. When applying tACS to modulate brain oscillations, the effect will however most likely not only be linear and will also influence other frequencies<sup>87</sup>. The mechanisms by which tACS can have an effect on motor function recovery in stroke patients, requires further investigation.

## **FUTURE DIRECTIONS IN UNDERSTANDING RECOVERY POST-STROKE**

### **Spontaneous neurobiological recovery in non-recoverers and recoverers**

In addition to investigate what influences the trajectory of spontaneous neurobiological recovery, we need to find out what prevents recovery of function to take place at all in a specific group of severely affected patients.

Measures of corticospinal tract integrity using transcranial magnetic stimulation (TMS) or diffusion tensor imaging (DTI) in the acute stage, reflecting white matter integrity or lesion load, show promise to predict motor outcome<sup>88-91</sup>. In particular, in patients with severe motor impairment in the first weeks post-stroke, these integrity measures show a large predictive capacity and likely reflect a conditional requirement for recovery to be able to take place at all<sup>14</sup>. Larger trials are needed to investigate the predictive capacity of cortical spinal tract integrity markers and determine for which groups of patients with various recovery patterns, other markers are required. Combining imaging and neurophysiological

measures of brain structure and function might be the key to understand the variability in spontaneous neurobiological recovery and response to therapies <sup>84,92,93</sup>.

### Optimizing trials in stroke rehabilitation

If we do not understand what influences spontaneous neurobiological recovery, it will be hard to establish which intervention can enhance it <sup>83,94</sup> or determine if a therapy can change non-recoverers into recoverers of spontaneous neurological recovery. Almost all phase III and IV trials in stroke rehabilitation failed to show superior treatment effects despite early promising findings in proof of concept studies. These neutral results may be related to our poor understanding of the interaction between reactive and experience-dependent plasticity during stroke recovery. Recommendation for future trials are given in Figure 8.1, see '*recommendations for (pre-) clinical effect studies*'. Acknowledging the small to moderate effect-sizes of effective training programs, future studies need to be more precise and based on better phenotyping of recovery. For this latter purpose, simple but robust markers are needed for selecting patients with and without the potential for spontaneous neurobiological recovery <sup>93,95,96</sup>. These fundamental research questions should be answered first to apply precise and effective treatment in the future <sup>97</sup>.

### Measuring behavioural restitution

To determine if and how behavioural restitution might be augmented by interventions, several steps are needed. The recommendations from stroke recovery and rehabilitation roundtable (SRRR) papers includes standardized use of clinical measurements following the International classification of functioning, disability, and health (ICF) model, comply with the COSMIN statements <sup>98</sup> and collect data at fixed times within clinical trials and cohort studies <sup>99,100</sup>. These recommendations were applied as much as possible in the current thesis; however, the currently available clinical measures might not be sufficient for understanding recovery of upper limb activities. Acknowledging that none of these clinical measurements are able to measure quality of movement, recently, the SRRR suggested a set of kinematic and kinetic measures that should be added to this core set of clinical measurements in order to better distinguish between behavioural restitution and compensation of function. The next challenge is to show interaction effects of neural repair with exercise therapy with or without support of neuromodulation or neuroprotective agents <sup>69,70</sup>.

### **Towards patient-specific prediction of recovery**

The current longitudinal studies only include patients with first-ever ischemic lesion and no severe comorbidities that could hamper recovery after stroke, which adheres to only a small subset of the clinical population. To predict individual recovery for the whole heterogeneous population of stroke patients, a large amount of data is needed for which standardized measuring is required. The collection of this data needs to go beyond research settings and should be conducted by all professionals in stroke rehabilitation. The Zon-Mw funded study, *Precision profiling to improve prognosis post-stroke: PROFITS* (ZonMw-funded Profits project (no 104003014), aims to collect a large dataset with repeated within-subject measurements. With that, PROFITS is a multicentre programme applied within stroke services aimed to improve precision profiling post-stroke by building a smart care chain. These kinds of consortium studies are needed to make prediction models of post-stroke recovery possible in the clinic and applicable for each individual patient.

## REFERENCES

- 1 Bolognini N, Russo C, Edwards DJ. The sensory side of post-stroke motor rehabilitation. *Restor Neurol Neurosci* 2016; **34**: 571–586.
- 2 Blennerhassett JM, Matyas TA, Carey LM. Impaired discrimination of surface friction contributes to pinch grip deficit after stroke. *Neurorehabil Neural Repair* 2007; **21**: 263–272.
- 3 Tyson SF, Hanley M, Chillala J, Selley AB, Tallis RC. Sensory loss in hospital-admitted people with stroke: characteristics, associated factors, and relationship with function. *Neurorehabil Neural Repair* 2008; **22**: 166–172.
- 4 Meyer S, De Bruyn N, Lafosse C, Van Dijk M, Michielsen M, Thijs L et al. Somatosensory impairments in the upper limb poststroke: distribution and association with motor function and visuospatial neglect. *Neurorehabil Neural Repair* 2016; **30**: 731–742.
- 5 Carey LM, Matyas TA, Baum C. Effects of somatosensory impairment on participation after stroke. *Am J Occup Ther* 2018; **72**: 1–10.
- 6 Twisk JWR, de Vente W. Hybrid models were found to be very elegant to disentangle longitudinal within- and between-subject relationships. *J Clin Epidemiol* 2019; **107**: 66–70.
- 7 Veerbeek, Winters C, Van Wegen EEH, Kwakkel G. Is the proportional recovery rule applicable to the lower limb after a first-ever ischemic stroke? *PLoS One* 2018; **13**: e0189279.
- 8 Turville ML, Matyas TA, Blennerhassett JM, Carey LM. Initial severity of somatosensory impairment influences response to upper limb sensory retraining post-stroke. *NeuroRehabilitation* 2018; **43**: 413–423.
- 9 Winters C, Van Wegen EEH, Daffertshofer A, Kwakkel G. Generalizability of the maximum proportional recovery rule to visuospatial neglect early poststroke. *Neurorehabil Neural Repair* 2017; **31**: 334–342.
- 10 Marchi NA, Ptak R, Di Pietro M, Schnider A, Guggisberg AG. Principles of proportional recovery after stroke generalize to neglect and aphasia. *Eur J Neurol* 2017; **24**: 1084–1087.
- 11 Winters C, Van Wegen EEH, Daffertshofer A, Kwakkel G. Generalizability of the proportional recovery model for the upper extremity after an ischemic stroke. *Neurorehabil Neural Repair* 2015; **29**: 614–622.
- 12 Prabhakaran S, Zarahn E, Riley C, Speizer A, Chong JY, Lazar RM et al. Inter-individual variability in the capacity for motor recovery after ischemic stroke. *Neurorehabil Neural Repair* 2008; **22**: 64–71.
- 13 Lazar RM, Minzer B, Antoniello D, Festa JR, Krakauer JW, Marshall RS. Improvement in aphasia scores after stroke is well predicted by initial severity. *Stroke* 2010; **41**: 1485–1488.
- 14 Byblow WD, Stinear CM, Barber PA, Petoe MA, Ackerley SJ. Proportional recovery after stroke depends on corticomotor integrity. *Ann Neurol* 2015; **78**: 848–859.
- 15 Hope TMH, Friston K, Price CJ, Leff AP, Rotshtein P, Bowman H. Recovery after stroke: not so proportional after all? *Brain* 2019; **142**: 15–22.
- 16 Kundert R, Goldsmith J, Veerbeek JM, Krakauer JW, Luft AR. What the proportional recovery rule is (and is not): methodological and statistical considerations. *Neurorehabil Neural Repair* 2019; **33**: 875–875.
- 17 van der Vliet R, Selles RW, Andrinopoulou ER, Nijland R, Ribbers GM, Frens MA et al. Predicting Upper Limb Motor Impairment Recovery after Stroke: A Mixture Model. *Ann Neurol* 2020; **87**: 383–393.
- 18 Nakayama H, Jorgensen HS, Raaschou HO, Olsen TS. The influence of age on stroke outcome. The Copenhagen Stroke Study. *Stroke* 1994; **25**: 808–813.
- 19 Bentsen L, Christensen L, Christensen A, Christensen H. Outcome and risk factors presented in old patients above 80 years of age versus younger patients after ischemic stroke. *J Stroke Cerebrovasc Dis* 2014; **23**: 1944–1948.
- 20 Stolk-Hornsveld F, Crow JL, Hendriks EP, Van Der Baan R, Harmeling-van Der Wel BC. The Erasmus MC modifications to the (revised) Nottingham Sensory Assessment: a reliable somatosensory assessment measure for patients with intracranial disorders. *Clin Rehabil* 2006; **20**: 160–172.
- 21 Connell LA, Tyson SF. Measures of sensation in neurological conditions: a systematic review. *Clin Rehabil* 2012; **26**: 68–80.



- 22 Kalogianni K, Saes M, Vlaar MP, Wegen EE. van, Kwakkel G, Schouten AC et al. Are longitudinal SSEP recordings a biomarker for proportional motor recovery post stroke? PhD thesis, Benefits and pitfalls in the longitudinal assessment of the somatosensory cortex post-stroke using EEG, <https://doi.org/10.4233/uuid:2dceae5b-145f-41fd-9b08-200d1e4781af>
- 23 Zandvliet SB, van Wegen EEH, Campfens SF, van der Kooij H, Kwakkel G, Meskers CGM. Position-cortical coherence as a marker of afferent pathway integrity early post-stroke, a prospective cohort study. *Neurorehabil Neural Repair* 2020; **34**: 344–359.
- 24 Campfens SF, Zandvliet SB, Meskers CGM, Schouten AC, van Putten MJAM, van der Kooij H. Poor motor function is associated with reduced sensory processing after stroke. *Exp Brain Res* 2015; **233**.
- 25 Rabiller G, He JW, Nishijima Y, Wong A, Liu J. Perturbation of brain oscillations after ischemic stroke: A potential biomarker for post-stroke function and therapy. *Int J Mol Sci* 2015; **16**: 25605–25640.
- 26 Finnigan S, van Putten MJAM. EEG in ischaemic stroke: Quantitative EEG can uniquely inform (sub-)acute prognoses and clinical management. *Clin Neurophysiol* 2013; **124**: 10–19.
- 27 Burghaus L, Hilker R, Dohmen C, Bosche B, Winhuisen L, Galdiks N et al. Early electroencephalography in acute ischemic stroke: Prediction of a malignant course? *Clin Neurol Neurosurg* 2007; **109**: 45–49.
- 28 Sheorajpanday RVA, Nagels G, Weeren AJTM, van Putten MJAM, De Deyn PP. Quantitative EEG in ischemic stroke: correlation with functional status after 6 months. *Clin Neurophysiol* 2011; **122**: 874–883.
- 29 Zhang S, Ke Z, Li L, Yip S, Tong K. EEG patterns from acute to chronic stroke phases in focal cerebral ischemic rats: correlations with functional recovery. *Physiol Meas* 2013; **34**: 423–435.
- 30 Machado C, Cuspineda E, Valdes P, Virues T, Llopis F, Bosch J et al. Assessing acute middle cerebral artery ischemic stroke by quantitative electric tomography. *Clin EEG Neurosci* 2004; **35**: 116–124.
- 31 Cuspineda E, Machado C, Galán L, Aubert E, Alvarez M a, Llopis F et al. QEEG prognostic value in acute stroke. *Clin EEG Neurosci* 2007; **38**: 155–160.
- 32 Finnigan SP, Rose SE, Walsh M, Griffin M, Janke AL, McMahon KL et al. Correlation of quantitative EEG in acute ischemic stroke with 30-day NIHSS score: comparison with diffusion and perfusion MRI. *Stroke* 2004; **35**: 899–903.
- 33 Laaksonen K, Helle L, Parkkonen L, Kirveskari E, Mäkelä JP, Mustanoja S et al. Alterations in spontaneous brain oscillations during stroke recovery. *PLoS One* 2013; **8**: e61146.
- 34 Gloor P, Ball G, Schaul N. Brain lesions that produce delta waves in the EEG. *Neurology* 1977; **27**: 326–333.
- 35 Ward NS. Using oscillations to understand recovery after stroke. *Brain* 2015; **138**: 2811–2813.
- 36 Nicolo P, Rizk S, Magnin C, Pietro M Di, Schnider A, Guggisberg AG. Coherent neural oscillations predict future motor and language improvement after stroke. *Brain* 2015; **138**: 3048–3060.
- 37 Wang Y, Zhang X, Huang J, Zhu M, Guan Q, Liu C. Associations between EEG beta power abnormality and diagnosis in cognitive impairment post cerebral infarcts. *J Mol Neurosci* 2013; **49**: 632–638.
- 38 Yang Y, Solis-Escalante T, van der Helm FCT, Schouten AC. A Generalized Coherence Framework for Detecting and Characterizing Nonlinear Interactions in the Nervous System. *IEEE Trans Biomed Eng* 2016; **63**: 2629–2637.
- 39 Vlaar MP, Birpoutsoukis G, Lataire J, Schoukens M, Schouten AC, Schoukens J et al. Modeling the Nonlinear Cortical Response in EEG Evoked by Wrist Joint Manipulation. *IEEE Trans Neural Syst Rehabil Eng* 2018; **26**: 205–215.
- 40 Yamada K, Mori S, Nakamura H, Ito H, Kizu O, Shiga K et al. Fiber-tracking method reveals sensorimotor pathway involvement in stroke patients. *Stroke* 2003; **34**: E159–62.
- 41 Schaechter JD, Moore CI, Connell BD, Rosen BR, Dijkhuizen RM. Structural and functional plasticity in the somatosensory cortex of chronic stroke patients. *Brain* 2006; **129**: 2722–2733.
- 42 Bocconi L, Meyer S, Kessner SS, De Bruyn N, Essers B, Cheng B et al. Is there full or proportional somatosensory recovery in the upper limb after stroke? Investigating behavioral outcome and neural correlates. *Neurorehabil Neural Repair* 2018; **32**: 691–700.


- 43 Lefebvre S, Liew S-L. Anatomical parameters of tDCS to modulate the motor system after stroke: a review. *Front Neurol* 2017; **8**: 29.
- 44 De Aguiar V, Paolazzi CL, Miceli G. tDCS in post-stroke aphasia: the role of stimulation parameters, behavioral treatment and patient characteristics. *Cortex*. 2015; **63**: 296–316.
- 45 Hsu TY, Juan CH, Tseng P. Individual differences and state-dependent responses in transcranial Direct Current Stimulation. *Front Hum Neurosci* 2016; **10**: 643.
- 46 Thirugnanasambandam N, Grundey J, Adam K, Drees A, Skwirba AC, Lang N et al. Nicotinic impact on focal and non-focal neuroplasticity induced by non-invasive brain stimulation in non-smoking humans. *Neuropsychopharmacology* 2011; **36**: 879–886.
- 47 Li LM, Uehara K, Hanakawa T. The contribution of interindividual factors to variability of response in transcranial direct current stimulation studies. *Front Cell Neurosci* 2015; **9**: 181.
- 48 Wiethoff S, Hamada M, Rothwell JC. Variability in response to transcranial direct current stimulation of the motor cortex. *Brain Stimul* 2014; **7**: 468–475.
- 49 Silvanto J, Muggleton N, Walsh V. State-dependency in brain stimulation studies of perception and cognition. *Trends Cogn Sci* 2008; **12**: 447–454.
- 50 Al-Kaysi AM, Al-Ani A, Loo CK, Breakspear M, Boonstra TW. Predicting brain stimulation treatment outcomes of depressed patients through the classification of EEG oscillations. In: *38th Annual International Conference of the IEEE Engineering in Medicine and Biology Society*. 2016, pp 5266–5269.
- 51 Stagg CJ, Antal A, Nitsche MA. Physiology of transcranial Direct Current Stimulation. *J ECT* 2018; **34**: 144–152.
- 52 Jackson MP, Rahman A, Lafon B, Kronberg G, Ling D, Parra LC et al. Animal models of transcranial direct current stimulation: Methods and mechanisms. *Clin Neurophysiol* 2016; **127**: 3425–3454.
- 53 Jamil A, Batsikadze G, Kuo HI, Labruna L, Hasan A, Paulus W et al. Systematic evaluation of the impact of stimulation intensity on neuroplastic after-effects induced by transcranial direct current stimulation. *J Physiol* 2017; **595**: 1273–1288.
- 54 Elsner B, Kugler J, Mehrholz J. Transcranial direct current stimulation (tDCS) for upper limb rehabilitation after stroke: future directions. *J. Neuroeng. Rehabil.* 2018; **15**: 106.
- 55 Hunter MA, Coffman BA, Trumbo MC, Clark VP. Tracking the neuroplastic changes associated with transcranial direct current stimulation: a push for multimodal imaging. *Front Hum Neurosci* 2013; **7**: 495.
- 56 Schonewille M, Belmeguenai A, Koekkoek SK, Houtman SH, Boele HJ, van Beugen BJ et al. Purkinje cell-specific knockout of the protein phosphatase PP2B impairs potentiation and cerebellar motor learning. *Neuron* 2010; **67**: 618–628.
- 57 Schonewille M, Gao Z, Boele HJ, Vinuesa Veloz MF, Amerika WE, Simek AAM et al. Reevaluating the role of LTD in cerebellar motor learning. *Neuron* 2011; **70**: 43–50.
- 58 D'Angelo E. The organization of plasticity in the cerebellar cortex: from synapses to control. *Prog Brain Res* 2014; **210**: 31–58.
- 59 Cooke SF, Bliss TVP. Plasticity in the human central nervous system. *Brain* 2006; **129**: 1659–73.
- 60 Jayaram G, Galea JM, Bastian AJ, Celnik P. Human locomotor adaptive learning is proportional to depression of cerebellar excitability. *Cereb Cortex* 2011; **21**: 1901–1909.
- 61 Medina JF, Lisberger SG. Links from complex spikes to local plasticity and motor learning in the cerebellum of awake-behaving monkeys. *Nat Neurosci* 2008; **11**: 1185–1192.
- 62 Ito M, Yamaguchi K, Nagao S, Yamazaki T. Long-Term Depression as a model of cerebellar plasticity. *Prog Brain Res* 2014; **210**: 1–30.
- 63 Galea JM, Jayaram G, Ajagbe L, Celnik P. Modulation of cerebellar excitability by polarity-specific noninvasive direct current stimulation. *J Neurosci* 2009; **29**: 9115–9122.
- 64 Galea JM, Vazquez A, Pasricha N, Orban De Xivry JJ, Celnik P. Dissociating the roles of the cerebellum and motor cortex during adaptive learning: The motor cortex retains what the cerebellum learns. *Cereb Cortex* 2011; **21**: 1761–1770.
- 65 Ferrucci R, Brunoni AR, Parazzini M, Vergari M, Rossi E, Fumagalli M et al. Modulating Human Procedural Learning by Cerebellar Transcranial Direct Current Stimulation. *The Cerebellum* 2013; **12**: 485–492.

- 66 Das S, Spoor M, Sibindi TM, Holland P, Schonewille M, De Zeeuw CI et al. Impairment of Long-Term Plasticity of Cerebellar Purkinje Cells Eliminates the Effect of Anodal Direct Current Stimulation on Vestibulo-Ocular Reflex Habituation. *Front Neurosci* 2017; **11**: 444.
- 67 Buch ER, Santarnecchi E, Antal A, Born J, Celnik PA, Classen J et al. Effects of tDCS on motor learning and memory formation: A consensus and critical position paper. *Clin Neurophysiol* 2017; **128**: 589–603.
- 68 Ferrucci R, Cortese F, Priori A. Cerebellar tDCS: How to Do It. *Cerebellum*. 2015. doi:10.1007/s12311-014-0599-7.
- 69 Bernhardt J, Borschmann KN, Kwakkel G, Burridge JH, Eng JJ, Walker MF et al. Setting the scene for the Second Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2019; **14**: 450–456.
- 70 Kwakkel G, Van Wegen E, Burridge JH, Winstein CJ, Van Dokkum L, Alt Murphy M et al. Standardized measurement of quality of upper limb movement after stroke: Consensus-based core recommendations from the Second Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2019; **14**: 783–791.
- 71 Fritsch B, Reis J, Martinowich K, Schambra HM, Ji Y, Cohen LG et al. Direct current stimulation promotes BDNF-dependent synaptic plasticity: Potential implications for motor learning. *Neuron* 2010; **66**: 198–204.
- 72 Murphy TH, Corbett D. Plasticity during stroke recovery: from synapse to behaviour. *Nat Rev Neurosci* 2009; **10**: 861–872.
- 73 Boddington LJ, Reynolds JNJ. Targeting interhemispheric inhibition with neuromodulation to enhance stroke rehabilitation. *Brain Stimul* 2017; **10**: 214–222.
- 74 Murase N, Duque J, Mazzocchio R, Cohen LG. Influence of Interhemispheric Interactions on Motor Function in Chronic Stroke. *Ann Neurol* 2004; **55**: 400–409.
- 75 Lindenberg R, Renga V, Zhu LL, Nair D, Schlaug G. Bihemispheric brain stimulation facilitates motor recovery in chronic stroke patients. *Neurology* 2010; **75**: 2176–2184.
- 76 Di Pino G, Pellegrino G, Assenza G, Capone F, Ferreri F, Formica D et al. Modulation of brain plasticity in stroke: a novel model for neurorehabilitation. *Nat Rev Neurol* 2014; **10**: 597–608.
- 77 Xu J, Branscheidt M, Schambra H, Steiner L, Widmer M, Diedrichsen J et al. Rethinking interhemispheric imbalance as a target for stroke neurorehabilitation. *Ann Neurol* 2019; **85**: 502–513.
- 78 Stinear CM, Petoe MA, Byblow WD. Primary Motor Cortex Excitability During Recovery After Stroke: Implications for Neuromodulation. *Brain Stimul* 2015; **8**: 1183–1190.
- 79 Sobesky J, Thiel A, Ghaemi M, Hilker RH, Rudolf J, Jacobs AH et al. Crossed cerebellar diaschisis in acute human stroke: a PET study of serial changes and response to supratentorial reperfusion. *J Cereb Blood Flow Metab* 2005; **25**: 1685–1691.
- 80 Wessel MJ, Hummel FC. Non-invasive cerebellar stimulation: a promising approach for stroke recovery? *Cerebellum* 2018; **17**: 359–371.
- 81 Carrera E, Tononi G. Diaschisis: past, present, future. *Brain* 2014; **137**: 2408–2422.
- 82 Bernhardt J, Borschmann K, Boyd L, Carmichael ST, Corbett D, Cramer SC et al. Moving Rehabilitation Research Forward: Developing Consensus Statements for Rehabilitation and Recovery Research. *Neurorehabil Neural Repair* 2017; **31**: 694–698.
- 83 Cramer SC. Repairing the human brain after stroke: I. Mechanisms of spontaneous recovery. *Ann. Neurol.* 2008; **63**: 272–287.
- 84 Ward NS. Restoring brain function after stroke - bridging the gap between animals and humans. *Nat Rev Neurol* 2017; **13**: 244–255.
- 85 Antal A, Paulus W. Transcranial alternating current stimulation (tACS). *Front Hum Neurosci* 2013; **7**: 317.
- 86 Herrmann CS, Rach S, Neuling T, Struber D. Transcranial alternating current stimulation: a review of the underlying mechanisms and modulation of cognitive processes. *Front Hum Neurosci* 2013; **7**: 279.
- 87 Antal A, Herrmann CS. Transcranial Alternating Current and Random Noise stimulation: possible mechanisms. *Neural Plast* 2016; **2016**: 3616807.

- 88 Bigourdan A, Munsch F, Coupe P, Guttmann CRG, Sagnier S, Renou P et al. Early fiber number ratio is a surrogate of corticospinal tract integrity and predicts motor recovery after stroke. *Stroke* 2016; **47**: 1053–1059.
- 89 Feng W, Wang J, Chhatbar PY, Doughty C, Landsittel D, Lioutas V-A et al. Corticospinal tract lesion load: An imaging biomarker for stroke motor outcomes. *Ann Neurol* 2015; **78**: 860–870.
- 90 Stinear CM, Barber PA, Petoe M, Anwar S, Byblow WD. The PREP algorithm predicts potential for upper limb recovery after stroke. *Brain* 2012; **135**: 2527–2535.
- 91 Talelli P, Greenwood RJ, Rothwell JC. Arm function after stroke: neurophysiological correlates and recovery mechanisms assessed by transcranial magnetic stimulation. *Clin Neurophysiol* 2006; **117**: 1641–1659.
- 92 Burke Quinlan E, Dodakian L, See J, McKenzie A, Le V, Wojnowicz M et al. Neural function, injury, and stroke subtype predict treatment gains after stroke. *Ann Neurol* 2015; **77**: 132–145.
- 93 Boyd LA, Hayward KS, Ward NS, Stinear CM, Rosso C, Fisher RJ et al. Biomarkers of stroke recovery: consensus-based core recommendations from the stroke recovery and rehabilitation roundtable. *Int J Stroke* 2017; **12**: 480–493.
- 94 Bernhardt J, Hayward KS, Kwakkel G, Ward NS, Wolf SL, Borschmann K et al. Agreed definitions and a shared vision for new standards in stroke recovery research: The Stroke Recovery and Rehabilitation Roundtable taskforce. *Int J Stroke* 2017; **12**: 444–450.
- 95 Ward NS. Does neuroimaging help to deliver better recovery of movement after stroke? *Curr Opin Neurol* 2015; **28**: 323–329.
- 96 Winters C, Heymans MW, Van Wegen EEH, Kwakkel G. How to design clinical rehabilitation trials for the upper paretic limb early post stroke? *Trials* 2016; **17**: 468.
- 97 Winters C, Kwakkel G, Van Wegen EEH, Nijland RHM, Veerbeek JM, Meskers CGM. Moving stroke rehabilitation forward: the need to change research. *NeuroRehabilitation* 2018; **43**: 19–30.
- 98 Mokkink LB, Terwee CB, Knol DL, Stratford PW, Alonso J, Patrick DL et al. The COSMIN checklist for evaluating the methodological quality of studies on measurement properties: A clarification of its content. *BMC Med Res Methodol* 2010; **10**: 22.
- 99 Kwakkel G, Lannin NA, Borschmann K, English C, Ali M, Churilov L et al. Standardized measurement of sensorimotor recovery in stroke trials: Consensus-based core recommendations from the Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2017; **12**: 451–461.
- 100 Bernhardt J, Kwakkel G, Lannin NA, Borschmann K, English C, Ali M et al. Consensus Statements from the Stroke Recovery and Rehabilitation Roundtable standardized measurement of sensorimotor recovery in stroke trials: Consensus-based core recommendations from the Stroke Recovery and Rehabilitation Roundtable. *J Stroke* 2017; **31**: 784–792.







## English summary

Spontaneous neurobiological  
recovery and modulation of  
sensorimotor function after  
ischemic stroke





Cerebral ischemic stroke accounts for about 87% of all stroke incidents in most Western countries<sup>1</sup>. An ischemic stroke leads to unilateral loss of function, called hemiparesis, in about 75% of patients<sup>2</sup>. Most patients remain affected in terms of body function, such as speech, motor, somatosensory and cognitive functions, as well as on an activity and participation level, such as motor activities, mobility, self-care, communication and social participation<sup>3</sup>. Task-specific and individualized rehabilitation strategies are needed to optimize meaningful activities in daily living such as standing balance, gait, and upper limb activities<sup>4</sup>. This thesis focusses on the underlying neuronal mechanisms of recovery post-stroke.

### **Neuronal mechanisms underlying recovery after stroke**

The majority of patients with a hemiparesis show some form of recovery in the first weeks after the stroke<sup>5</sup>. Based on animal studies, there is strong evidence that several mechanisms are responsible for observed true neurological repair in the brain. These mechanisms are a response to a sudden reduced regional cerebral blood flow caused by a thrombus or embolus in one of the main arteries of the brain and referred to as reactive plasticity within this thesis. The time window of the cascade of hemodynamic and neuroinflammatory reactions starts already within minutes<sup>6-8</sup> and may extend to several months. Some of these neuroinflammatory reactions such as tonic GABA-ergic inhibition of anatomically related, non-infarcted, areas are suppressive beyond the first weeks, whereas other reactions are enhancing brain plasticity within the first weeks after stroke onset<sup>7,9</sup>. Due to the optimal conditions for enhanced plasticity in this specific early time window of 8–10 weeks after the stroke, this period is also considered as the optimal time window for intervention to promote recovery. In contrast to animal studies, it is unknown if and how true neurological repair can be influenced in humans<sup>10</sup>. Brain plasticity not only occurs as a reaction after the stroke, it takes place throughout the human lifespan, and can strengthen or weaken specific functions<sup>11,12</sup>. This process of learning allows us to adapt to the requirements of the environment and is known as experience-dependent plasticity. Both these non-learning and learning dependent mechanisms of plasticity are important in post-stroke rehabilitation<sup>10</sup>.

### **Spontaneous neurobiological recovery after stroke**

Due to the above described mechanisms of plasticity in the brain, stroke patients can show neurological recovery<sup>7</sup>. This spontaneous neurobiological recovery seems to be proportional to the severity of the initial paresis of the upper limb within the first 72 hours after the stroke<sup>13,14</sup>. Recent studies suggest that, for the majority of people, this so-called proportional recovery is between 50 and 90% of the maximal possible recovery on the Fugl-Meyer motor assessment of the upper extremity (FM-UE)<sup>13,14</sup>. While the majority of

patients show at least some level of motor recovery of the upper limb, this is not the case for about one third of patients<sup>14</sup>. An accurate individual prediction of a patient's potential for recovery is unfortunately not yet possible. Individual prediction of recovery is needed for clinical care, as well as for patient selection in clinical trials to develop the optimal treatment for different subsets of patients<sup>15</sup>.

The first chapters of this thesis concern observational studies and focus on non-learning-dependent mechanisms which can contribute to improvement in motor function after stroke. Potential clinical and neurophysiological biomarkers for recovery of motor function were monitored with repeated measurements in time in the first 6 months after stroke.

**Chapter 2** focused on the influence of somatosensory impairment on arm-hand motor recovery in the first 6 months after stroke. In 94 patients with motor and somatosensory impairments, measured with the FM-UE and Erasmus modification of the Nottingham sensory assessment (EmNSA), in the first 3 weeks after the stroke, the recovery for both modalities were measured repeatedly over time.

We investigated if the relationship between motor and somatosensory recovery could reflect a parallel recovery in both modalities in the first 6 months post-stroke and if intactness of somatosensory function is a pre-requisite for upper limb motor recovery early post-stroke. The results showed that the relationship between the two modalities can only partially be explained by global mechanisms of spontaneous neurobiological recovery. Since the association between motor and somatosensory recovery for patients with severe somatosensory impairment at baseline did not significantly differ from the whole group, we concluded that somatosensory impairment at baseline does not prevent motor recovery. In contrast, we found significant differences in the association between motor and somatosensory recovery when comparing patients who show motor recovery (recoverers) and patients who show little to no motor recovery (non-recoverers), separately. In patients who had a mild motor impairment or showed an improvement of 6 points or more on the FM-UE at 26 weeks post-stroke (recoverers), motor recovery was found to be positively associated with somatosensory recovery, independent of progress of time. In addition to mechanisms that drive spontaneous neurobiological recovery in both modalities, recovery of somatosensory impairment seems conditional for full motor recovery.

Clinical measures provide information on patients' degree of recovery and can partially predict how a person will recover in the following months. Unfortunately, these measures have so far not made it possible to accurately predict the recovery of each individual patient. Markers of brain activity, measured prior to functional recovery might potentially be beneficial in improving prediction models and could help us to better understand the

underlying mechanisms in the brain. For example, neurophysiological markers of brain activity, measured using electroencephalography (EEG), could yield possible markers for spontaneous neurobiological recovery.

### Neurophysiological markers of brain activity

In resting state EEG, increased power of slow oscillations are consistently found after stroke and correlates with stroke severity and a larger infarction volume<sup>16,17</sup>. In **chapter 3**, EEG was longitudinally measured in 41 patients during awake rest to evaluate spectral characteristics as a representation of neuronal deficits and their development over time in relation to recovery of motor function and stroke severity. In agreement with literature<sup>18</sup>, we also found increased power in the low frequency delta band of the affected hemisphere. The power in the delta band was represented relative to alpha activity, i.e. delta-alpha ratio of the affected hemisphere ( $DAR_{AH}$ ), and relative to the unaffected hemisphere, i.e. directional BSI ( $BSI_{dir_{delta}}$ ).  $DAR_{AH}$  and  $BSI_{dir_{delta}}$  were associated with stroke severity, measured with the national institutes of health stroke scale, within 3 weeks post-stroke. Stroke induced neural tissue alterations are likely responsible for this decrease in frequency of neuronal oscillations.  $DAR_{AH}$  and  $BSI_{dir_{delta}}$  showed potential as biomarkers in future prediction models, reflecting individual spontaneous neurobiological recovery and can potentially distinguish between patients with different recovery patterns, so called recoverers and non-recoverers. However, resting-state spectral characteristics which quantify the change in frequencies of the neural oscillations seem to be a rather global measure of pathology in the brain. Our study suggests that in order to capture a patient's potential for spontaneous neurobiological recovery, a specific focus on the sensorimotor system itself is required. For patients with severe upper limb impairments with absence of voluntary motor control, such a task can obviously only be passive.

In **chapters 4 and 5**, system identification techniques were used to investigate how passive wrist joint manipulations are transferred to the brain. Using a robotic arm, the wrist was passively moved in a pattern with specific frequencies, while EEG activity was simultaneously measured. The similarity between both signals was calculated as the position-cortical coherence (PCC) as a measure of somatosensory pathway integrity. Since somatosensory function is important for movement control, it could be a possible marker for motor recovery after stroke. A previous study showed that PCC can be reliably measured in healthy subjects<sup>19</sup>. **Chapter 4** examined whether this is also the case for people with a stroke. All 11 measured patients showed significant contralateral PCC. A relationship was found between PCC and motor impairment, measured with the FM-UE. These results led to the longitudinal study described in **chapter 5**, in which PCC was

measured serially in 48 patients to capture the dynamics of PCC in the time window of spontaneous neurobiological recovery. The amount of electrodes and frequencies, on which PCC was significantly measured (%PCC), increased from baseline to 12 weeks post-stroke, which was in accordance with the recovery seen on the EmNSA and FM-UE scales. A significant positive association was found between %PCC, mean amplitude in the affective hemisphere (Amp-A) and EmNSA of the hand and fingers. PCC was also higher at baseline in terms of percentage and amplitude in patients who showed motor recovery at 26 weeks post-stroke as compared to patients with poor or no recovery. Our results demonstrated the longitudinal construct validity of %PCC and Amp-A as a measure of somatosensory pathway integrity. A high %PCC in recoverers with a low baseline FM-UE however suggests that this measure also contains information on cortical excitability. More advanced computational approaches are needed to determine if PCC has a merit as a marker for spontaneous neurobiological recovery of somatosensory function.

### **Modulation of sensorimotor recovery after stroke**

Next to understanding spontaneous neurobiological recovery after stroke, a key challenge is to optimize experience-dependent plasticity and ultimately understand if reactive and experience-dependent plasticity can influence each other to enhance recovery. The second part of this thesis focussed on learning-dependent mechanisms for recovery. **Chapters 6 and 7** aimed to investigate the potential to modulate sensorimotor recovery of standing balance performance by transcranial direct current stimulation (tDCS).

#### *Transcranial direct current stimulation*

In transcranial direct current stimulation (tDCS) a weak direct current (up to 3 mA) flows between electrodes placed onto the skull. tDCS is assumed to induce polarity specific alterations in the membrane potential of neurons, thereby increasing neuronal excitability in the brain in the area of stimulation<sup>20,21</sup>. Cathodal stimulation will generally induce low and anodal stimulation will generally induce high concentration of intracellular calcium, leading to respectively long term depression (LTD) or long term potentiation (LTP) like plasticity<sup>21</sup>. tDCS has been extensively investigated as a tool to optimize rehabilitation after stroke, in which the majority of studies have focused on the upper limb<sup>22,23</sup>. A current meta-analysis indicates a low to moderate quality of evidence for improving ADL performance with tDCS applied to the motor cortex after stroke and no evidence for the effectiveness of tDCS to enhance recovery of the upper and lower limb<sup>24</sup>.

The meta-analysis performed by Marquez et al. indicated that while no added value of tDCS was found on a group level, significant improvements were found when only patients

with a low to moderate level of impairment, were taken into account <sup>25</sup>. tDCS research therefore needs to focus on those patients in which tDCS can have an added value.

### ***Cerebellar tDCS***

The cerebellum contains about 80% of all neurons in the human brain <sup>26</sup>. Recently, there has been increased attention for the possible beneficial effects of cerebellar (cb)-tDCS in post-stroke motor function recovery as an alternative for cortical stimulation. The cerebellum could potentially be a target for stimulation due to its important function in error-based learning <sup>27–30</sup>.

In **chapter 6** of this thesis, standing balance performance was investigated as a target for enhanced experience-dependent plasticity by cb-tDCS. A group of 15 patients with a chronic stroke received anodal cb-tDCS on the ipsilesional cerebellar hemisphere, contralateral cerebellar hemisphere and sham stimulation in separate sessions while performing a dynamic postural feedback-based tracking task. After receiving ipsilesional cb-tDCS, we found an instantaneous positive effect on a composite centre of pressure measure of stability of standing balance, but only in the most challenging tandem stance position.

Although changes in balance performance were observed at group level in **chapter 6**, high interindividual variability in response to cb-tDCS was found, which indicates that the exact mechanisms by which cb-tDCS enforces its effects are not clear. In order to fully take advantage of the potentially enhancing effects of tDCS on experience-dependent plasticity by LTP and LTD-like mechanisms, it is essential to have thorough evidence and understanding of the mechanisms by which tDCS can enforce its effects and from that point of view explain and understand variability.

### **Modulation of sensorimotor recovery in the time window of spontaneous neurobiological recovery by cb-tDCS**

In the first 4–10 weeks post-stroke, neurotrophic factors create a critical time window for recovery. This unique time window of heightened plasticity early after stroke might give a unique chance to induce clinically relevant effects, which are unattained by therapy alone. If tDCS is able to accelerate experience-dependent plasticity, this effect has to be evident in this specific time window of enhanced homeostatic plasticity. Longitudinal studies that combine behavioural with neurophysiological and imaging measures, starting in the acute phase after stroke, could give valuable insight into the cause and effect of motor recovery after stroke and the mechanisms by which tDCS enforces its effects.

The randomized double-blind and sham-controlled POTENTIAL trial, of which the protocol is described in **chapter 7**, aims to determine if cb-tDCS applied in the early phase after stroke can enhance experience-dependent plasticity in the time window of spontaneous neurobiological recovery. The effects of tDCS are studied by a combination of clinical, neurophysiological and posturographical measurements to learn more about the underlying mechanisms by which cb-tDCS enforces its effects. Combining behavioural outcome measures with measures of functional brain network changes will enable us to relate hypothesized mechanisms of action of enhanced experience-dependent plasticity in the time window of spontaneous neurobiological recovery.

### **Future directions in understanding recovery of function post-stroke**

To determine if and how behavioural restitution might be enhanced by interventions, several steps are needed. Recommendations for future research are given in the general discussion in **chapter 8**.

Without understanding what drives spontaneous neurobiological recovery, it will be hard to establish whether and how it can be enhanced<sup>31,32</sup> or determine if a therapy can change non-recoverers into recoverers of spontaneous neurological recovery. Previous neutral results in large phase three trials may be related to our poor understanding of the interaction between reactive and experience-dependent plasticity during stroke recovery. Future studies need to be more precise and based on better phenotyping of recovery. For this latter purpose, simple but robust prognostic markers are needed for selecting patients with and without a potential for spontaneous neurobiological recovery<sup>33-35</sup>. Next to that, the currently available clinical measures might not be sufficient for understanding recovery of upper limb function. To measure quality of movement that is assumed to be mainly driven by behavioural restitution, a set of kinematic and kinetic measures is advised in order to better distinguish behavioural restitution from compensation of function<sup>36</sup>.

In addition, to investigate what influences the trajectories of spontaneous neurobiological recovery, we need to find out which factors prevent recovery of function to take place at all in a specific group of severely affected patients. Combining neuroimaging and neurophysiological measures of brain structure and function might be the key to understand the variability in spontaneous neurobiological recovery and response to therapies<sup>8,33,37</sup>. These fundamental research questions should be answered for precise understanding of spontaneous neurobiological recovery and factors that influences its trajectory. This knowledge is needed to be able to design and develop interventions that can enhance the speed of spontaneous neurological recovery and/or change non-recoverers into recoverers after stroke<sup>15</sup>.

## REFERENCES

- 1 Mozaffarian D, Benjamin EJ, Go AS, Arnett DK, Blaha MJ, Cushman M et al. Executive summary: heart disease and stroke statistics--2016 update: a report from the american heart association. *Circulation* 2016; **133**: 447–454.
- 2 Lawrence ES, Coshall C, Dundas R, Stewart J, Rudd AG, Howard R et al. Estimates of the prevalence of acute stroke impairments and disability in a multiethnic population. *Stroke* 2001; **32**: 1279–1284.
- 3 World Health organization. International Classification of Functioning, Disability and Health. Geneva, 2001.
- 4 Langhorne P, Bernhardt J, Kwakkel G. Stroke rehabilitation. *Lancet* 2011; **377**: 1693–1702.
- 5 Kwakkel G, Kollen B, Lindeman E. Understanding the pattern of functional recovery after stroke: facts and theories. *Restor Neurol Neurosci* 2004; **22**: 281–299.
- 6 Buma F, Kwakkel G, Ramsey N. Understanding upper limb recovery after stroke. *Restor Neurol Neurosci* 2013; **31**: 707–722.
- 7 Murphy TH, Corbett D. Plasticity during stroke recovery: from synapse to behaviour. *Nat Rev Neurosci* 2009; **10**: 861–872.
- 8 Ward NS. Restoring brain function after stroke - bridging the gap between animals and humans. *Nat Rev Neurol* 2017; **13**: 244–255.
- 9 Krakauer JW, Carmichael ST. *Broken Movement*. 2019 doi:10.7551/mitpress/9310.001.0001.
- 10 Zeiler SR, Krakauer JW. The interaction between training and plasticity in the poststroke brain. *Curr Opin Neurol* 2013; **26**: 609–616.
- 11 Cooke SF, Bliss TVP. Plasticity in the human central nervous system. *Brain* 2006; **129**: 1659–1673.
- 12 Feldman DE, Brecht M. Map plasticity in somatosensory cortex. *Science* 2005; **310**: 810–815.
- 13 Prabhakaran S, Zarahn E, Riley C, Speizer A, Chong JY, Lazar RM et al. Inter-individual variability in the capacity for motor recovery after ischemic stroke. *Neurorehabil Neural Repair* 2008; **22**: 64–71.
- 14 Winters C, Van Wegen EEH, Daffertshofer A, Kwakkel G. Generalizability of the proportional recovery model for the upper extremity after an ischemic stroke. *Neurorehabil Neural Repair* 2015; **29**: 614–622.
- 15 Winters C, Kwakkel G, Van Wegen EEH, Nijland RHM, Veerbeek JM, Meskers CGM. Moving stroke rehabilitation forward: the need to change research. *NeuroRehabilitation* 2018; **43**: 19–30.
- 16 Finnigan S, van Putten MJAM. EEG in ischaemic stroke: Quantitative EEG can uniquely inform (sub-)acute prognoses and clinical management. *Clin Neurophysiol* 2013; **124**: 10–19.
- 17 Burghaus L, Hilker R, Dohmen C, Bosche B, Winhuisen L, Galdiks N et al. Early electroencephalography in acute ischemic stroke: Prediction of a malignant course? *Clin Neurol Neurosurg* 2007; **109**: 45–49.
- 18 Gloor P, Ball G, Schaul N. Brain lesions that produce delta waves in the EEG. *Neurology* 1977; **27**: 326–333.
- 19 Campfens SF, Schouten AC, van Putten MJAM, van der Kooij H. Quantifying connectivity via efferent and afferent pathways in motor control using coherence measures and joint position perturbations. *Exp brain Res* 2013; **c**: 141–153.
- 20 Nitsche MA, Paulus W. Excitability changes induced in the human motor cortex by weak transcranial direct current stimulation. *J Physiol* 2000; **527**: 633–639.
- 21 Stagg CJ, Antal A, Nitsche MA. Physiology of transcranial Direct Current Stimulation. *J ECT* 2018; **34**: 144–152.
- 22 Elsner B, Kugler J, Pohl M, Mehrholz J. Transcranial direct current stimulation (tDCS) for improving aphasia in patients after stroke. *Cochrane database Syst Rev* 2013; CD009760.
- 23 Elsner B, Kugler J, Pohl M, Mehrholz J. Transcranial direct current stimulation (tDCS) for improving activities of daily living, and physical and cognitive functioning, in people after stroke. *Cochrane database Syst Rev* 2016; **3**: CD009645.



- 24 Elsner B, Kwakkel G, Kugler J, Mehrholz J. Transcranial direct current stimulation (tDCS) for improving capacity in activities and arm function after stroke: a network meta-analysis of randomised controlled trials. *J Neuroeng Rehabil* 2017; **14**: 95.
- 25 Marquez J, Van Vliet P, Mcelduff P, Lagopoulos J, Parsons M. Transcranial direct current stimulation (tDCS): Does it have merit in stroke rehabilitation? A systematic review. *Int. J. Stroke*. 2015; **10**: 306–316.
- 26 Azevedo FAC, Carvalho LRB, Grinberg LT, Farfel JM, Ferretti REL, Leite REP *et al*. Equal numbers of neuronal and nonneuronal cells make the human brain an isometrically scaled-up primate brain. *J Comp Neurol* 2009; **513**: 532–541.
- 27 D'Angelo E. The organization of plasticity in the cerebellar cortex: from synapses to control. *Prog Brain Res* 2014; **210**: 31–58.
- 28 Medina JF, Lisberger SG. Links from complex spikes to local plasticity and motor learning in the cerebellum of awake-behaving monkeys. *Nat Neurosci* 2008; **11**: 1185–1192.
- 29 Schlerf J, Ivry RB, Diedrichsen J. Encoding of sensory prediction errors in the human cerebellum. *J Neurosci* 2012; **32**: 4913–4922.
- 30 Pollok B, Butz M, Gross J, Südmeyer M, Timmermann L, Schnitzler A. Coupling between cerebellar hemispheres: Behavioural, anatomic, and functional data. *Cerebellum* 2006; **5**: 212–219.
- 31 Bernhardt J, Hayward KS, Kwakkel G, Ward NS, Wolf SL, Borschmann K *et al*. Agreed definitions and a shared vision for new standards in stroke recovery research: The Stroke Recovery and Rehabilitation Roundtable taskforce. *Int J Stroke* 2017; **12**: 444–450.
- 32 Cramer SC. Repairing the human brain after stroke: I. Mechanisms of spontaneous recovery. *Ann. Neurol*. 2008; **63**: 272–287.
- 33 Boyd LA, Hayward KS, Ward NS, Stinear CM, Rosso C, Fisher RJ *et al*. Biomarkers of stroke recovery: consensus-based core recommendations from the stroke recovery and rehabilitation roundtable. *Int J Stroke* 2017; **12**: 480–493.
- 34 Ward NS. Does neuroimaging help to deliver better recovery of movement after stroke? *Curr Opin Neurol* 2015; **28**: 323–329.
- 35 Winters C, Heymans MW, Van Wegen EEH, Kwakkel G. How to design clinical rehabilitation trials for the upper paretic limb early post stroke? *Trials* 2016; **17**: 468.
- 36 Kwakkel G, Van Wegen E, Burridge JH, Winstein CJ, Van Dokkum L, Alt Murphy M *et al*. Standardized measurement of quality of upper limb movement after stroke: Consensus-based core recommendations from the Second Stroke Recovery and Rehabilitation Roundtable. *Int J Stroke* 2019; **14**: 783–791.
- 37 Burke Quinlan E, Dodakian L, See J, McKenzie A, Le V, Wojnowicz M *et al*. Neural function, injury, and stroke subtype predict treatment gains after stroke. *Ann Neurol* 2015; **77**: 132–145.







## Nederlandse samenvatting

Spontaan neurobiologisch  
herstel en modulatie van  
sensomotorische functie  
na een beroerte





Een cerebro vasculair accident (CVA) ofwel beroerte, wordt veroorzaakt door een blokkade van de bloedtoevoer naar een hersengebied (herseninfect) dan wel door een bloeding (hersensbloeding). Het in dit proefschrift beschreven onderzoek richt zich voornamelijk op patiënten met een herseninfect, ongeveer 87% van de patiënten met een beroerte. Een herseninfect kan leiden tot uitvalsverschijnselen of spierzwakte van een lichaamshelft, ook wel (hemi-)parese genoemd, maar kan ook spraak-, taal- en andere cognitieve functiestoornissen tot gevolg hebben. Een parese komt naar schatting in 75% van de patiënten met een herseninfect voor. Een groot deel van deze patiënten blijft langdurig beperkt in hun functioneren, gemeten op 6 tot 12 maanden na de beroerte.

### **Onderliggende processen tijdens het herstel na een beroerte**

In de eerste weken na de beroerte herstellen de hersenen van een 'shock'. Weefsel wat te weinig zuurstof heeft gekregen maar niet irreversibel is beschadigd kan zich in die periode herstellen. Ook hersengebieden die op een afstand in verbinding stonden met het geïnfarceerde gebied, kunnen zich herstellen waarbij ook nieuwe netwerken zich kunnen vormen. Aangenomen wordt dat deze veranderingen kunnen plaatsvinden door, onder meer, een verhoogde afgifte van groeihormonen die zorgen dat hersenen zich kunnen aanpassen. Deze neurologische veranderingen worden binnen dit proefschrift reactieve plasticiteit genoemd en kunnen leiden tot een aanzienlijke verbetering in de neurologische functie in de eerste weken na de beroerte.

Eerder onderzoek heeft laten zien dat een groot deel van de patiënten herstel van de parese laat zien. Dit herstel bereikt een plateau in de eerste 4 tot 10 weken na de beroerte. Bovendien lijkt het herstel in verhouding te staan tot de ernst van de parese vlak na de beroerte. Recente onderzoeken suggereren dat dit zogenoemde proportionele motorisch herstel van de parese voor de meeste patiënten tussen de 50 en 90% ligt van het herstel wat maximaal mogelijk is op de betreffende klinische meetschaal. Helaas laat niet iedereen herstel zien. Een kwart tot ongeveer één derde van de patiënten herstelt niet tot nauwelijks en blijft een zwaar aangedane arm- en handfunctie houden. Voor andere modaliteiten, zoals beenfunctie of verminderde ruimtelijke aandacht, is hetzelfde proportioneel herstel gerapporteerd waarbij meestal ook een subpopulatie is gevonden die niet tot nauwelijks herstelt. Deze gemeten verbetering, mede als gevolg van reactieve plasticiteit, wordt in dit proefschrift spontaan neurobiologisch herstel genoemd en lijkt hetzelfde beloop te hebben ongeacht de modaliteit.

Naast de hierboven beschreven reactieve plasticiteit als gevolg van de beroerte kunnen ook andere vormen van hersensplasticiteit bijdragen aan het herstel na een beroerte. Neuronale

veranderingen in de hersenen treden gedurende de gehele levensduur van de mens op en kunnen specifieke functies versterken of verzwakken. Dit proces van leerafhankelijke plasticiteit is, naast reactieve plasticiteit, belangrijk in de revalidatie na een beroerte. Of leerafhankelijke plasticiteit ook reactieve plasticiteit kan beïnvloeden is nog niet bekend.

### **Doel van dit proefschrift**

Binnen de in dit proefschrift beschreven studies is onderzoek gedaan naar spontaan neurobiologisch herstel, voorspellers van dit proces vroeg na de beroerte en factoren die dit proces negatief of positief kunnen beïnvloeden. Om therapieën effectief en efficiënt toe te passen en een realistische verwachting van het herstel en het leven met een beroerte te geven, is het belangrijk goed te kunnen voorspellen of en hoeveel iemand neurologisch en functioneel zal herstellen. Momenteel kan men nog niet voor elke individuele patiënt voorspellen of hij of zij zal verbeteren. Daarnaast is nog nauwelijks bekend welke factoren spontaan neurobiologisch herstel positief of negatief beïnvloeden en of bepaalde therapieën herstel kunnen verhogen of versnellen.

De eerste hoofdstukken van dit proefschrift betreffen observationele studies waarin het verloop van spontaan neurobiologisch herstel wordt beschreven in het eerste half jaar na de beroerte. Daarbij is gekeken naar voorspellers en mogelijke belangrijke factoren voor het motorische herstel van de arm en hand. In het tweede deel van dit proefschrift is gekeken of leerafhankelijke plasticiteit versterkt kan worden door middel van non-invasieve hersenstimulatie. Daarnaast wordt het protocol beschreven van een gerandomiseerde studie naar het effect van non-invasieve hersenstimulatie op stabalans in de vroege fase na een beroerte. Door een combinatie van hersenstimulatie tijdens stabalanstraining al vroeg na een beroerte te geven kan onderzocht worden of deze combinatietherapie een meerwaarde heeft door leerafhankelijke plasticiteit te stimuleren in de tijdsperiode waarbinnen ook spontaan neurobiologisch herstel plaats kan vinden.

### **Spontaan neurobiologisch herstel na een beroerte**

In **hoofdstuk 2** is gekeken naar stoornissen in de waarneming van tast, pijn, temperatuur en het gevoel voor houding en beweging na de beroerte, samen somatosensorische functiestoornissen genoemd. Hierbij is gekeken naar de invloed van somatosensorische functiestoornissen op het motorisch herstel van de arm en hand in de eerste 6 maanden na een beroerte. In een groep van 94 patiënten die in de eerste 3 weken na de beroerte zowel motorische als somatosensorische functiestoornissen van de arm en hand hadden is het herstel op beide domeinen herhaald in de tijd gemeten. In dit prospectieve onderzoek



is gekeken of het verbeteren van de functiestoornissen op beide domeinen verklaard zou kunnen worden door het domeinoverstijgende proces van spontaan neurobiologisch herstel. Daarnaast is gekeken of een somatosensorische functiestoornis een mogelijke beperkende invloed heeft op het motorisch herstel van de arm en hand. De resultaten lieten zien dat de relatie tussen beide domeinen voor een belangrijk deel verklaard kan worden door een generiek domeinoverstijgend proces van spontaan neurobiologisch herstel. Binnen de onderzochte populatie had een ernstige somatosensorische functiestoornis niet per se een remmende werking op motorisch herstel. Wel was het niet of slecht herstellen van de somatosensorische functiestoornis in de maanden na de beroerte gerelateerd aan een verminderd motorisch herstel. Concluderend lijkt het herstel van somatosensorische functiestoornissen voorwaardelijk te zijn voor volledig motorisch herstel en is het belangrijk om beide functiestoornissen te monitoren vroeg na een beroerte.

Klinische meetschalen geven een beeld van de mate van herstel van een patiënt. Sommige meetschalen, kort na de beroerte of herhaaldelijk gemeten, geven informatie over het verwachte verloop van het herstel en kunnen gedeeltelijk voorspellen hoe iemand gaat herstellen in de daaropvolgende maanden. Om herstel beter te begrijpen moet gezocht worden naar markers in de hersenen die voorspellend zijn voor spontaan neurobiologisch herstel. Hersenactiviteit gemeten met behulp van elektro-encefalografie (EEG) zou mogelijke markers voor spontaan neurobiologisch herstel kunnen opleveren. In hoofdstuk 3 t/m 5 van dit proefschrift is onderzoek gedaan naar deze mogelijke markers vanuit het EEG.

Voor de onderzoeken in **hoofdstuk 3 en 5** zijn patiënten gedurende het eerste half jaar na de beroerte gevolgd om het herstel van de arm- en handfunctie in kaart te brengen en prospectief te onderzoeken of dit herstel gerelateerd is aan maten vanuit het EEG. Patiënten zijn voor dit onderzoek binnen de eerste 3 weken en vervolgens op 5, 12 en 26 weken na de beroerte gemeten. Deze metingen vonden plaats in een speciaal ingerichte meetbus met benodigde apparatuur om metingen op locatie te kunnen uitvoeren. Hierdoor konden patiënten worden onderzocht zonder dat de patiënt hiervoor hoefde te reizen. Naast EEG werden verschillende klinische meetschalen herhaaldelijk afgenomen om het herstel van elke patiënt over de tijd te kunnen volgen.

**Hoofdstuk 3** beschrijft de ontwikkeling van verschillende EEG-maten over de tijd bij 41 patiënten met een primair herseninfarct. EEG werd gemeten tijdens rust met de ogen open. In overeenstemming met eerdere onderzoeken werd bij patiënten een verhoogde delta-activiteit (frequentieband 1–4 Hz) gevonden in de aangedane hersenhelft. De EEG-maten normaliseerden geleidelijk over de eerste 6 maanden, ook voorbij 12 weken na de beroerte, waardoor het patroon niet volledig overeen kwam met het spontaan neurobiologisch herstel gemeten met de klinische maten. De verhouding tussen delta-

activiteit in de aangedane en niet-aangedane hersenhelft ( $BSI_{\text{delta}}$ ) en de verhouding tussen delta- en alpha-activiteit (frequentieband 7–12 Hz) in de aangedane hersenhelft ( $DAR_{\text{AH}}$ ), waren gerelateerd aan verbeteringen in globaal neurologische stoornissen, gemeten met de National Institutes of Health stroke scale (NIHSS). alleen  $BSI_{\text{delta}}$  was gerelateerd aan het motorisch herstel, gemeten met de Fugl-Meyer motor assessment van de arm (FM-UE). Concluderend lijkt rust EEG een indicatie te geven van de globale neurologische schade in de hersenen. Voor het voorspellen van motorisch herstel lijken specifiekere neurofysiologische maten nodig te zijn.

In **hoofdstuk 4 en 5** is onderzoek gedaan met behulp van een systeem identificatie techniek waarmee gekeken kan worden hoe het signaal van een mechanische beweging van de pols wordt verwerkt in de hersenen. Met behulp van een robotarm werd de pols passief bewogen in een patroon met specifieke frequenties; dit verstoringssignaal werd vervolgens vergeleken met gelijktijdig gemeten EEG-activiteit. De overeenkomst tussen beide signalen werd vervolgens berekend als de positie-corticale coherentie (PCC), de overeenkomst tussen de polsbeweging en het gemeten EEG-signaal. Aangezien deze maat voor signaaloverdracht gerelateerd is aan somatosensorische functie, wat belangrijk is voor de sturing van beweging, zou het ook een mogelijke marker kunnen zijn voor het te verwachten motorisch herstel vlak na een beroerte. Nadat eerder onderzoek heeft laten zien dat PCC betrouwbaar kan worden gemeten in gezonde proefpersonen, is in **hoofdstuk 4** geconstateerd dat dit ook het geval is bij patiënten met een beroerte. Ook werd gevonden dat de PCC-maten gerelateerd waren aan de motorische functie van patiënten met een beroerte. Deze resultaten gaven aanleiding tot de longitudinale studie in **hoofdstuk 5**, waarbij PCC gemeten is in 48 patiënten gedurende de eerste 6 maanden na de beroerte. Motorische functiestoornissen van de arm en hand werden gemeten met de FM-UE en somatosensorische functiestoornissen van de vinger en pols met de Erasmus MC modificatie van de Nottingham sensory assessment (EmNSA). In dit onderzoek kwam naar voren dat het patroon van de ontwikkeling van de PCC-maten over de tijd parallel verloopt met de snelle spontane verbetering in motorische en somatosensorische functiestoornissen, in de eerste weken na de beroerte. Deze stijgende lijn vlakkt vervolgens af tussen 12 en 26 weken na de beroerte. Daarmee lijkt PCC, in ieder geval deels, het proces van spontaan neurobiologisch herstel te meten. De PCC-maten bleken gerelateerd te zijn aan somatosensorische functie maar niet aan motorische functie. In de groep patiënten die, ondanks een zeer zwaar aangedane motorische functie van de arm, in de eerste 3 weken na de beroerte later toch een goed herstel lieten zien, waren de gemeten PCC-waardes significant hoger dan in de groep die geen goed herstel van motorische functie liet zien. Dit resultaat lijkt er op te wijzen dat de hersenen een sterkere reactie geven op het verstoringssignaal in de groep patiënten die een groter herstel

lieten zien. Concluderend lijkt PCC niet alleen iets te zeggen over de intactheid van de signaaloverdracht tussen de pols en de hersenen, maar lijkt ook de sterkte van de reactie van hersenen informatief te kunnen zijn. Deze resultaten laten potentieel zien voor de toekomst maar kunnen in vervolgonderzoek nog verbeterd worden door geavanceerde methodes die de betrouwbaarheid van de meting zullen moeten verbeteren.

### **Modulatie van sensomotorisch herstel na een beroerte**

Aangezien stabilans belangrijk is voor veel activiteiten in het dagelijks leven, zoals lopen en het uitvoeren van transfers, is het streven dit zo snel mogelijk na de beroerte te verbeteren. Mogelijk zijn de hersenen direct na de beroerte gevoeliger en ontvankelijker voor leren en kan daarmee spontaan neurobiologisch herstel beïnvloed worden. In **hoofdstuk 6 en 7** is onderzocht of het verbeteren van stabilans na een beroerte versneld kan worden door tijdens een balustraining hersenstimulatie te geven. Transcraniële direct current stimulatie (tDCS) is een niet-invasieve techniek waarbij de neuronen onder de positief geladen elektroden in een verhoogde staat van paraatheid gebracht kunnen worden. Het idee achter deze hersenstimulatie is dat als de neuronen tijdens het uitvoeren van een taak sneller vuren, men daardoor sneller een taak zou kunnen leren. In dierexperimenteel onderzoek zijn na het toepassen van tDCS groeihormonen gemeten die gerelateerd zijn aan leerprocessen. Mogelijk kan deze techniek het (her)leren van activiteiten of vaardigheden in de revalidatie versnellen of mogelijk zelfs spontaan neurobiologisch herstel beïnvloeden. De kleine hersenen (het cerebellum) spelen een belangrijke rol bij de controle van de stabilans en tijdens het leren van een nieuwe taak. Mits de beroerte niet het cerebellum heeft getroffen zou het optimaliseren van de processen in het cerebellum door middel van een combinatie van training en tDCS een positief effect kunnen hebben op de stabilans bij patiënten met een beroerte. In **hoofdstuk 6** is onderzoek gedaan naar de korte termijneffecten van cerebellaire tDCS voor het verbeteren van de stabilans bij 15 patiënten die langer dan 6 maanden voor het onderzoek een beroerte hebben gehad maar nog steeds balansstoornissen hadden. Stabilans werd in dit onderzoek gemeten met behulp van een krachtenplaat waarmee schommelingen in de lichaamspositie gemeten kunnen worden tijdens staan, staan met de ogen dicht en staan met een voet voor de andere (tandemstand). De resultaten lieten zien dat patiënten na elke trainingssessie stabiel stonden in de tandemstand. In de trainingssessie waarbij de hersenstimulatie gegeven werd aan de zijde van het cerebellum die verbonden is met het aangedane been was de meeste vooruitgang in stabilans zichtbaar. Voor het bewerkstelligen van een merkbare verbetering in het dagelijks leven van patiënten is meer dan een enkele trainingssessie nodig. Om dit te onderzoeken is een gerandomiseerde en geblindeerde


effectstudie opgezet beschreven in **hoofdstuk 7**. In deze studie werd gekeken wat het effect is van een intensieve balanstraining van meerdere weken, toegepast in de eerste weken na de beroerte. Hierbij zijn klinische meetschalen gecombineerd met metingen met behulp van een krachtplaat en EEG. Door deze combinatie van technieken kan niet alleen gekeken worden naar het effect van deze gecombineerde training, maar ook meer inzicht gekregen worden in de onderliggende processen van spontaan neurobiologisch herstel.

In de algemene discussie in **hoofdstuk 8** wordt geconcludeerd dat toekomstig onderzoek zich zal moeten richten op het verder in kaart brengen van de voorspelbaarheid en beïnvloedbaarheid van spontaan neurobiologisch herstel. Hierin moet een onderscheid gemaakt worden in factoren die iets zeggen over de ernst van de beroerte, bijvoorbeeld de NIHSS-score vlak na de beroerte, en factoren die meerwaarde hebben om het daadwerkelijke herstel in de eerste 6 maanden na de beroerte in kaart te brengen, bijvoorbeeld de verbetering van motorische functie gemeten met de FM-UE. Om het effect van revalidatie-interventies goed te kunnen meten zijn gerandomiseerde effectstudies nodig waarbij in de opzet van het onderzoek rekening gehouden moet worden met het spontane neurobiologische herstel. Het spontane neurobiologische herstelproces zorgt immers in een specifieke groep patiënten voor verbeteringen, terwijl dit in anderen achterblijft. Toekomstig onderzoek naar de meerwaarde van vroeg ingezette interventies zal daarom duidelijk onderscheid moeten maken tussen het mogelijke effect van een interventie en het eventuele spontane neurologische herstel. Dit kan gedaan worden door hypothesen te richten op een specifieke subgroep van patiënten die bij aanvang zowel in tijd als ernst prognostisch vergelijkbaar zijn en daarmee kansrijk zijn om een eventueel interactie-effect te vertonen tussen enerzijds het spontane neurobiologische herstel en anderzijds therapie. Deze selectie van een homogene groep dient plaats te vinden samen met een randomisatieprocedure. Om interactie-effecten te kunnen meten zullen naast klinische ook technologische meetinstrumenten gebruikt moeten worden die in staat zijn om kwaliteit van bewegen te kunnen meten. Deze aanbevelingen zijn in lijn met de recente consensus van experts in het onderzoeksveld en zullen er in de toekomst voor moeten zorgen dat patiënten met een beroerte therapie op maat gegeven kan worden.









Dankwoord  
(Acknowledgements)





Wat zijn er veel mensen die mij geholpen of gesteund hebben bij het schrijven van dit proefschrift.

Allereerst wil ik alle deelnemers bedanken voor het meewerken aan de onderzoeken. Voor velen al heel kort na een beroerte, in een heftige tijd waarin opeens je hele leven op zijn kop staat. Zonder jullie belangeloze hulp zou dit proefschrift er niet zijn.

Hooggeleerde en/of zeer geleerde opponenten, prof. dr. Geert Verheyden, prof. dr. Sander Geurts, prof. dr. Michel van Putten, prof. dr. Maarten Frens, dr. Marieke Visser en Prof. dr. Frans Nollet, hartelijk dank voor het beoordelen van mijn proefschrift. Ik kijk ernaar uit met jullie, online of offline, in discussie te gaan.

Hooggeleerde prof. dr. Kwakkel en zeer geleerde dr. van Wegen en dr. Meskers, beste Gert, Erwin en Carel, dank voor het vertrouwen dat ik van jullie heb gekregen om dit traject te kunnen starten. Jullie hebben mij als masterstudent en beginnend promovendus, in het LUMC en daarna in het VUmc, geholpen om het best mogelijke uit mezelf en de onderzoeken te halen. Gert, dank voor je passie en gedrevenheid voor het vakgebied en dat je ondanks je drukke agenda altijd tijd voor me maakte. Daarnaast wil ik je ook bedanken voor je hulp om verder te gaan in de wetenschap en het contact dat je voor me hebt gelegd in Zwitserland. Erwin, dank voor je positiviteit, vertrouwen en dat je deur altijd open stond om even te sparren of een praatje te maken. Carel, dank voor je snelle en scherpe blik. Voor alle ingewikkelde problemen wisten we op de vrijdagochtenden in 't Refpunt toch altijd weer oplossingen te bedenken.

Daarnaast wil ik alle medeauteurs bedanken, Rinske Nijland, Mique Saes, Aukje Andringa, Andreas Daffertshofer, Jos Twisk, Floor Campfens, Alfred Schouten, Michel van Putten en Herman van der Kooij. Dank voor jullie kritische blikken, tips, feedback en hulp bij het tot stand komen van de hoofdstukken.

Floor, wat is het al weer lang geleden dat ik je voor het eerst kwam helpen bij je metingen in het LUMC. Heel fijn dat je me toen alles kon vertellen over het bestaan als promovendus en dat ik later het EXPLORE-stroke onderzoek van je over kon nemen, dank je wel.

Voor het uitvoeren van de onderzoeken ben ik altijd hartelijk ontvangen bij Reade, Vogellanden en Revant. Binnen deze instellingen en ook in het VUmc zijn er veel mensen die me hebben geholpen bij de opzet, organisatie en uitvoering van de onderzoeken. In het bijzonder wil ik Rinske, Maurits en Marianne bedanken voor jullie hulp met de organisatie van het POTENTIAL onderzoek binnen Reade en Vogellanden. Rosa, Jonnie, Ellen, Marijke, Sonja, Betty, Marijn, Quirine, Marianne, Anna, Louise, Chermène, Yvonne, Martin, Jos en Emmy, dank voor het geven van de therapie binnen de studies en/of hulp

bij de werving van deelnemers. Mique, Luuk, Dirk, Elza, Aukje, Caroline, Inez en Natasja en de vele studenten, dank voor jullie hulp bij de metingen en de gezellige ritjes in de meetbus. Edwin, dank voor het gereedmaken van de meetbus. Hans, wat heb ik veel van je geleerd, en wat heb je me vaak geholpen bij problemen met apparatuur. Ivan, wat was het fijn dat je onze kamer zó gezellig vond dat je soms binnen een paar minuten voor mijn neus stond om een computerprobleem te verhelpen. Anja, dank voor je hulp en gezelligheid, ik wens je veel sterkte en gezondheid toe.

Alle fijne collega's van de afdeling Revalidatiegeneeskunde hebben er aan bijgedragen dat ik 5 jaar lang met veel plezier in het VUmc heb gewerkt. Ik kijk met plezier terug op de praatjes tijdens het thee zetten, zes trappen omhoog met de goedkope koffieclub, wandelingen rond de VU, muizen vangen, de lunch in het 't Refpunt, het jaarlijkse kerstontbijt en alle borrels. Janne en Marc, dank voor alle wijze adviezen, Janneke voor de aanmoediging en Lizanne voor het afspelen van een kattenfilmpje op het moment dat ik dat heel hard nodig had. Aukje, Caroline en Mique, bij jullie als kamergenootjes kon ik met alles terecht. Aukje en Caroline, door mijn verhuizing naar Zwitserland en Covid is ons laatste etentje al weer even geleden, maar hopelijk plannen we snel weer een volgende keer. Emma, laten we snel weer een WODje doen als ik in Nijmegen ben.

Dear prof. dr. Hummel, dear Friedhelm, thank you for giving me the opportunity to grow further as a researcher. Dear Hummellab colleagues, thank you for welcoming me in the lab family, I am looking forward to celebrate it with you.

Familie en vrienden, jullie bleven altijd geïnteresseerd in mijn onderzoek, ook al mochten jullie er van mij soms niet meer naar vragen. Dank voor alle steun en gezelligheid. Ik kijk er naar uit om jullie allemaal te knuffelen als dat weer kan.

Jorien en Caroline met jullie als lieve, wijze paranimfen heb ik zo veel zin in de dag.

Pap, ik weet dat je heel trots bent op je twee (bijna) gepromoveerde kinderen en dat mama dat ook geweest zou zijn.

Harmen, wat is het fijn om je naast me te hebben.









About the author  
List of publications





## ABOUT THE AUTHOR

Sarah Zandvliet was born on December 1<sup>st</sup>, 1987 in Venray. She attended secondary school at the Raayland college in Venray, where she graduated in 2005. From 2005 until 2009, Sarah studied Physiotherapy at the Utrecht University of Applied sciences (HU). She was immediately drawn to neurorehabilitation. During the third year she performed a five month internship in the outpatient rehabilitation centre in Värnamo, Sweden, specialized in acquired brain injuries and neuromuscular diseases. Her interest in research was sparked while writing her bachelor thesis on the fear of falling in stroke patient. In 2009 she obtained her bachelor degree and continued with a pre-qualification program in order to be admitted to the Research Master clinical and fundamental human movement sciences. During her master, she worked as a research assistant on a fall prevention project in older adults with prof. dr. Pijnappels. Sarah received her Master's degree Cum Laude in 2013, her thesis was titled: *Assessing afferent pathways to improve prediction of motor recovery after stroke*. The research on which her thesis was based was performed as part of the EXPLORE-stroke study in the Leiden university medical centre (LUMC). She was awarded with as university fellowship with prof. dr. Kwakkel for the academic year 2013–2014 and joined the team as a PhD student. She continued working on the EXPLORE-stroke project in the LUMC and later the Amsterdam university medical centre, and started the POTENTIAL project as part of the research described in this thesis.

Currently, Sarah works in the lab of prof. dr. Hummel at the Swiss Federal Institute of Technology Lausanne (EPFL), embedded in the rehabilitation centre in Sion and Biotech campus in Geneva. In this position she is working on several projects related to stroke recovery.

## LIST OF PUBLICATIONS

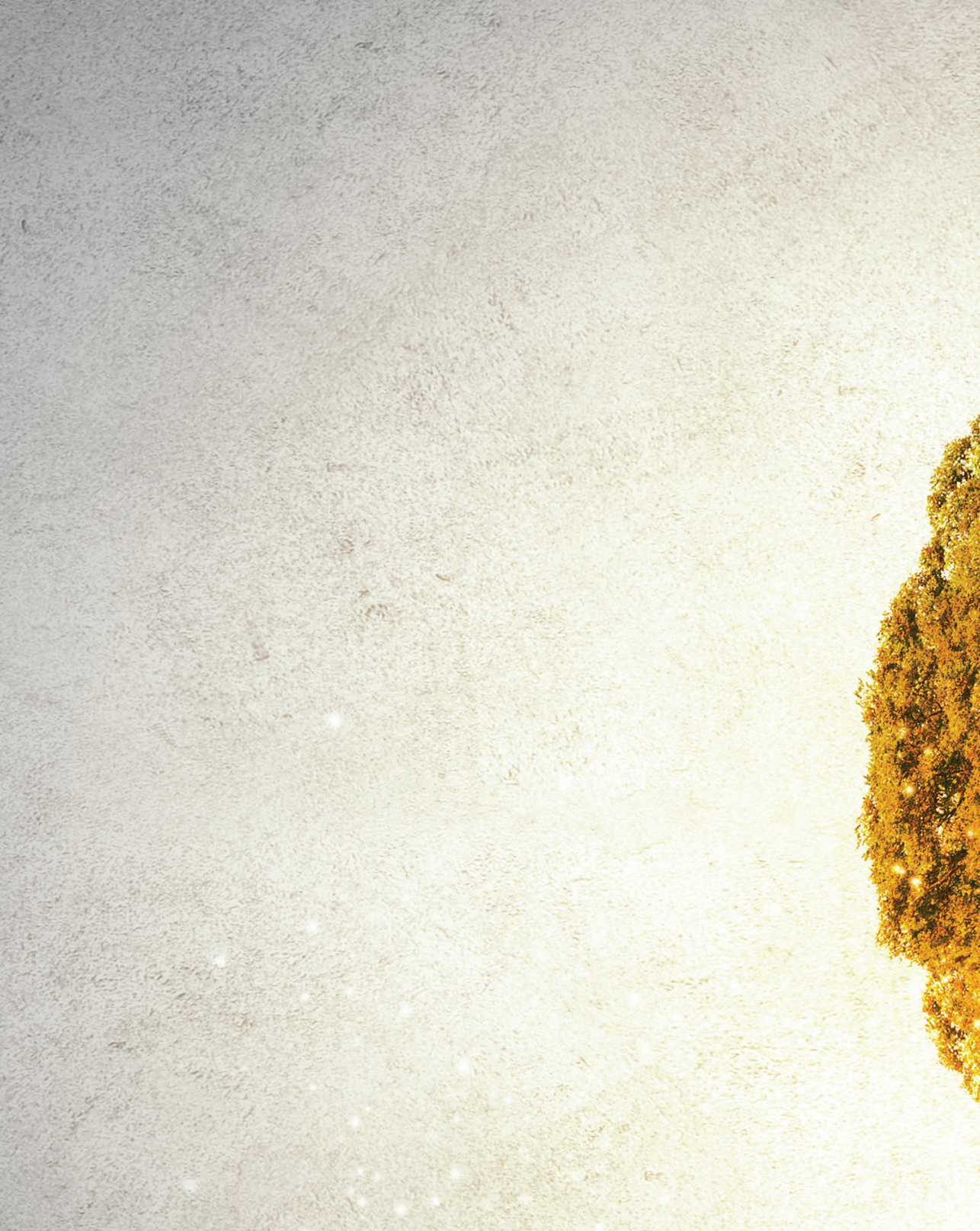
### Publications related to this thesis

- Is recovery of somatosensory impairment conditional for upper limb motor recovery after stroke? Sarah B. Zandvliet, Gert Kwakkel, Rinkse H.M. Nijland, Erwin, E.H. van Wegen, Carel G.M. Meskers, *Neurorehabilitation and Neural repair* 2020; **34**(5): 403–416.
- Is resting-state EEG longitudinally associated with spontaneous neurobiological recovery early post-stroke? A prospective cohort study, M. Seas\*, Sarah B. Zandvliet\*, Aukje S. Andringa, Andreas Daffertshofer, Jos W. R. Twisk, Carel G.M. Meskers, Erwin E.H. van Wegen, Gert Kwakkel, on behalf of the 4D-EEG consortium, *Neurorehabilitation and Neural repair* 2020; **34**(5): 389–402. \* Authors contributed equally.
- Position-cortical coherence as a marker of afferent pathway integrity early post-stroke: a prospective cohort study. Sarah B. Zandvliet, Erwin E.H. van Wegen, S. Floor Campfens, Herman van der Kooij, Gert Kwakkel, Carel G.M. Meskers, *Neurorehabilitation and Neural repair* 2020; **34**(4): 344–359.
- The effect of Cerebellar transcranial direct current stimulation to improve standing balance performance early post-stroke, study protocol of a randomized controlled trial. Sarah B. Zandvliet, Carel G.M. Meskers, Rinske H.M. Nijland, Andreas Daffertshofer, Gert Kwakkel, Erwin E.H. van Wegen, *International Journal of Stroke* 2019; **14**(6): 650–657.
- Short term effects of cerebellar tDCS on standing balance performance in patients with chronic stroke and healthy age matched elderly. Sarah B. Zandvliet, Carel G.M. Meskers, Gert Kwakkel, Erwin E.H. van Wegen, *the Cerebellum* 2018; **17**(5): 575–589.
- Poor motor function is associated with reduced sensory pathway integrity after stroke. Floor Campfens, Sarah B. Zandvliet, Carel Meskers, Alfred Schouten, Michiel van Putten, Herman van der Kooij, *Experimental Brain Research* 2015; **233**(4): 1339–1349.

### Other publications

- Het POTENTIAL onderzoek in Vogellanden: Virtuele balanstraining gecombineerd met transcraniële elektrische stimulatie van de hersenen bij revalidanten met balansproblemen na een CVA. Sarah Zandvliet, Carel Meskers, Gert Kwakkel, Erwin van Wegen. *Reflex Vogellanden* 2017.

- POTENTIAL: Stimulerende balanstherapie na een beroerte. **Sarah Zandvliet**, Carel Meskers, Rinske Nijland, Gert Kwakkel, Erwin van Wegen, *Nederlands tijdschrift voor Geriatrische fysiotherapie* 2016; 10: 39–42.
- Virtual reality balanstherapie na een beroerte, het Potential project. **Sarah Zandvliet**, Carel Meskers, Rinske Nijland, Gert Kwakkel, Erwin van Wegen, *Reade Magazine* 2016.
- The relation of brain activity and recovery of arm-hand function after stroke. Luuk Haring, Mique Saes, **Sarah Zandvliet**, Caroline Winters, Aukje Andringa, *Dutch magazine of rehabilitation medicine* 2016; 4: 79–81.
- Balanstherapie met virtual reality na een beroerte, het Potential project. **Sarah Zandvliet**, Carel Meskers, Rinske Nijland, Gert Kwakkel, Erwin van Wegen, *Synaps VUmc* 2013.
- Valangst na een beroerte. **Sarah Zandvliet**, *HBO kennisbank* 2009.



Amsterdam  
Movement  
Sciences



Amsterdam Movement Sciences conducts scientific research to optimize physical performance in health and disease based on a fundamental understanding of human movement in order to contribute to the fulfillment of a meaningful life.