MAX PLANCK INSTITUTE FOR HUMAN COGNITIVE AND BRAIN SCIENCES LEIPZIG

Smadar Ovadia-Caro: Plasticity following stroke: The recovery of functional networks as measured by resting-state functional connectivity. Leipzig: Max Planck Institute for Human Cognitive and Brain Sciences, 2016 (MPI Series in Human Cognitive and Brain Sciences; 179)

Plasticity following stroke: the recovery of functional networks as measured by resting-state functional connectivity

Impressum

Max Planck Institute for Human Cognitive and Brain Sciences, 2016

Druck: Sächsisches Druck- und Verlagshaus Direct World, Dresden

Titelbild: © Smarda Ovadia-Caro, 2016

ISBN 978-3-941504-65-3

Plasticity following stroke: the recovery of functional networks as measured by resting-state functional connectivity

Dissertation

zur Erlangung des akademischen Grades Doctor rerum naturalium (Dr. rer. nat.)
im Fach Psychologie

Eingereicht an der Lebenswissenschaftliche Fakultät
der Humboldt-Universität zu Berlin

von
M.Sc., Smadar A., Ovadia-Caro, Geburtsname: Ovadia
Geboren am 14.12.1979 in Tel-Aviv, Israel

Präsident der Humboldt-Universität zu Berlin
Prof. Dr. Jan-Hendirk Olbertz

Dekan der Lebenswissenschaftliche Fakultät
Prof. Dr. Elmar Kulke

Gutachterin/Gutachter:
Prof. Dr. Elke van der Meer
Prof. Dr. Arno Villringer
Prof. Dr. Ulrich Dirnagl

Tag der Verteidigung:
4th August 2016

"Any man could, if he were so inclined, be the sculptor of his own brain"

Ramón y Cajal, Santiago, 1852-1934
Advice for a Young Investigator

Acknowledgements

Science is not a single person's work. Although I see some interesting similarities between scientists and artists, I still think that the more effective way to conduct research is as a joint initiative with other fun, creative brains.

I find myself in writing these lines thinking of the past five years of my life, which were very interesting I must say. Coming to a new place, learning a new language (to some extent, there have been complaints), and floating in the cloud of scientific problems as Uri Alon puts it. I have also co-founded my own family here in the land of wonderful summers and horrible winters.

My path into science did not follow a predetermined chronology. I am one of these lucky people to whom the door to the world of science opened up almost unexpectedly. And I still think this is one of the most exciting professions the modern world can offer us. I am happy to say I have some talent in bumping into very nice, smart, helpful and interesting people. This made the whole PhD process possible and much more enjoyable.

I would firstly like to thank my supervisors. Prof. van der Meer for her calm guidance, providing clarity in both the scientific path and also in life. Thank you for all the fruitful discussions we had. I always felt relaxed, focused and motivated after our meetings. You taught me to accept things I cannot change as they are and keep focused to push forward all the rest.

My sincere thank you to Prof. Arno Villringer for his continuing support and encouragement. With every meeting opening up with the question "how's life?" caring and helping in any possible way to move the research forward is not just a cliché in your case. I learned a lot both scientifically as well as on managing people, and how to find their inner motivation point. You care for your students very much and the result is that, at least for me, I felt a great responsibility to make you proud.

I thank Daniel Margulies for supporting me in my first scientific steps in Berlin. Your true passion for science is simply contagious. It was at times a bit chaotic with analyses in the middle of the night, and meetings after sleepless nights but it was also funny and inspiring, and I have learned a great deal.

I would like to thank my collaborators at the Charité hospital, Jochen Fiebach and Jan Jungehülsing. A special thank you is reserved to Kersten Villringer who introduced me to the world of stroke radiology, answered all my questions with endless patience (and those of you who know me know that's a lot), collected, (and is still collecting) the data used for my research. Thank you for all your kind help.

I thank Gabriele Lohmann for the wonderful collaboration on the CCM project. Who would have ever thought that a mathematician (very special people) and a non-mathematician (all the rest) could get along so well? It was truly a pleasure to work with you, diving into formulas and talking about common methods used in the field with a new perspective in mind.

My work would not have been possible without the generous funding of the Minerva foundation and the Berlin School of Mind and Brain. I would like to thank Sieglinde Reichardt from the Minerva foundation for her friendly, kind assistance and Annette Winkelmann and the devoted staff at the Berlin School of Mind and brain for their

support. A special thank you to Anne Löchte for her tremendous care in issues concerning career (and personal) development. I would also like to thank Conny Ketscher and Birgit Mittag from the MPI in Leipzig for their ever-smiling faces and constant assistance.

To my colleagues and friends, I can't thank enough. Felix Hasler, Christiane Rohr and Isabelle Bareither, my roommates in the lab; lunchtime was never boring with you guys. A critical neuroscientist on the one side of the table and EEG/fMRI scientists on the other - man! That was funny. My friends and collaborators from Leipzig, Judy Kipping and Alex Schäfer – two very passionate scientists, who always had good advice on any question concerning data analysis, graph theory (as well as embroidery and craftwork - Judy). Guido Hesselmann, who functions as my scientific compass ever since my master's studies – will always have an image of you riding your rocket bike in the August heat on the empty inner road of the Weizmann Institute (some things that only a German can do). Ella Striem-Amit from the Hebrew University, also mother of twins, for the collaboration on the project of connectivity in the blind. It was a pleasure to work together scientifically but most importantly to form our own small support group of those who live in constant chaos due to very fortunate life circumstances. Ahmed Khalil, who recently joined our labs, for his refreshing, realistic perspectives on science - I really enjoy talking to you and working together. And finally, to Fatma Imamoglu, my close friend, for all the wonderful days in the tree house. Working, talking, cooking, baking and working and talking again in long weekends with sleepy cats on the window. Trying to solve, (or at least analyze thoroughly), every possible problem, scientific or other.

For reading and commenting, thereby making this dissertation a better manuscript, I would like to thank Fatma, Guido and Ahmed again. I thank Isabelle and Karin Ludwig for kindly agreeing to translate the summary into German.

If you have prevailed the longest acknowledgment section in the history of dissertations at the HU, you have reached the most important part. I would like to thank my parents, two lovely people, for their hard work and strong belief that I can do almost anything. Albeit unrealistic, their confidence had a tremendous influence on the core wiring of my developing brain, convincing me there is nothing wrong with exploring very diverse interests, as long as you really mean that.

To Nadav - my husband, my partner, my friend. Following my dreams and ideas, wherever they may lead us both cannot be taken for granted. I think very few partners are supportive of their other halves as you are. Being an amazing father to Jonathan and Netta, while solving bugs in very unorthodox hours of the night are only some of the tasks only you can manage so well. You always have so many questions (and some good advice) on all the topics close to my heart. I think this should become the revised textbook definition for love. Toda Raba.

Eidesstattliche Erklärung

Hiermit versichere ich, dass ich die vorliegende Arbeit selbständig und nur unter Zuhilfenahme der angegebenen Quellen verfasst und die wörtlich oder inhaltlich entnommenen Stellen aus den verwendeten Quellen als solche kenntlich gemacht habe.

Weiterhin erkläre ich, dass ich mich nicht anderwärts um einen Doktorgrad beworben habe, keinen Doktorgrad in dem Promotionsfach Psychologie besitze und dass ich die zugrunde liegende Promotionsordnung vom 17.01.2005 kenne.

Smadar Ovadia-Caro
Berlin, den 6 May 2016

Table of Contents

Summary

Stroke is defined as a neurological deficit caused by a sudden, localized injury to the central nervous system due to a vascular pathology**.** The traditional clinical approach for understanding the origin of stroke symptoms was to attribute them to the local infarct. However, regions interconnected with the infarct may also be implicated in the symptom manifestation and recovery process, even if they remain structurally intact after stroke – a theory put forward over a century ago as diaschisis. The theory suggests that remote interconnected areas undergo plastic changes and are indirectly impaired by the local infarct.

With the development of resting-state functional magnetic resonance imaging (rs-fMRI), it is now possible to explore alterations in functional connections between distant areas on the macroscale level using a single scan. This approach has been previously applied to study plasticity following stroke, with some promising results linking connectivity alterations with behavioral deficits. Based on these findings, novel therapeutic approaches, such as non-invasive brain stimulation techniques are aimed at modulating connectivity within the affected network.

Stroke poses an experimental challenge due to the heterogeneity of both lesion location and clinical symptoms. Previous studies were therefore focused on connectivity changes either based on similarity of lesion location or similarity of symptoms, restricting the analysis to one functional network and corresponding neurological deficit. However, it is rarely the case that two stroke lesions are identical and most importantly, stroke generally results in more than one functional deficit. For these reasons, a multi-network assessment of plasticity is needed.

It was therefore the main goal of the current dissertation to develop a methodological approach that explores plasticity after stroke at the multi-network level using rs-fMRI data. This enables the exploration of patients with heterogeneous lesions, providing the possibility of a unified model to study recovery processes following a multifaceted neurological damage at the individual level.

Study I presents a novel mathematical analysis to rs-fMRI data computing the reproducibility of functional connectivity patterns using the concordance correlation coefficient, a quantitative measure of spatial similarity. This method can be applied to longitudinal datasets acquired after stroke to detect areas demonstrating large alterations in the functional connectivity structure over time. Based on our findings in healthy controls, and previous studies linking functional connectivity changes and behavioral impairment after stroke, we suggest concordance as a functionally meaningful measure of plasticity as reflected in changes of the spatial pattern of connectivity maps.

Building upon the methodological tool presented in study I, study II introduces a model for multi-network assessment of functional connectivity after stroke and its application to a group of stroke patients with heterogeneous lesions. We

computed concordance at the network level to quantify changes in functional connectivity over time. We found that on the individual level, even in cases of multi-network damage, functional connectivity is preferentially altered over time in affected networks, demonstrating lower concordance. Importantly, the degree of change in functional connectivity over time was correlated with the behavioral trajectory. This study provides empirical evidence supporting the theoretical model of diaschisis and further generalizes results obtained in single networks, among them the sensory, motor, attention and default-mode, to multiple large-scale networks, regardless of the lesion location or implicated network.

Study III is a review paper aimed at exploring the recent trends in the rapidly evolving field of rs-fMRI after stroke. We provide a summary of studies published thus far and discuss the utility and limitations of the approach. Based on the reviewed literature we describe the potential usefulness of rs-fMRI to stroke diagnosis in the acute phase as well as to the study of reorganization and plasticity in the subacute and chronic stages. We describe the model of stroke as a network disruption, and present the shift in the different methodologies applied to the study of plasticity following stroke given this model.

To summarize, based on our findings from the three studies discussed in this dissertation, we suggest a conceptual and methodological framework to study stroke as a network disruption rather than a mere localized phenomenon. This view of stroke has experimental implications, and most importantly potential therapeutic value in forming individualized protocols for non-invasive stimulation techniques.

Zusammenfassung

Ein Schlaganfall wird definiert als akutes neurologisches Defizit, das durch eine vaskuläre Pathologie (Ischämie oder Blutung) entsteht. Traditionell versucht man, die Schlaganfall-Symptome als Folge einer umschriebenen fokalen Schädigung zu verstehen. Allerdings können auch weitere, mit den geschädigten Regionen verbundene Hirnareale die Symptomausprägung und den Heilungsprozess beeinflussen - selbst wenn sie strukturell intakt erscheinen. Die vor mehr als einem Jahrhundert begründete *Diaschisis*-Theorie besagt, dass entfernte, aber verbundene Areale plastischen Veränderungen unterzogen und indirekt durch den lokalen Infarkt beeinträchtigt werden können.

Mit der Entwicklung der sogenannten „resting-state" funktionellen Magnetresonanztomographie (rs-fMRT) ist es inzwischen möglich, Veränderungen in funktioneller Konnektivität zwischen weit entfernten Arealen mit einem einzigen Scan abzubilden. Frühere Studien nutzten diesen Ansatz bereits zur Untersuchung von Plastizität nach einem Schlaganfall und lieferten einige vielversprechende Resultate, welche den Zusammenhang zwischen Veränderungen der funktionellen Konnektivität und neurologischen Symptomen aufzeigten. Auf Grundlage dieser Resultate versuchen derzeit neue therapeutische Ansätze, wie etwa nicht-invasive Hirnstimulations-Techniken, die Konnektivität in betroffenen Netzwerken zu modulieren.

Ein zentrales Problem für die Untersuchung von Plastizität nach einem Schlaganfall ist die ausgeprägte Heterogenität des Läsionsortes wie auch die damit verbundene Vielfalt klinischer Symptome. In bisherigen Studien zur Konnektivität nach einem Schlaganfall konzentrierte man sich deswegen auf Schlaganfälle mit möglichst gleichartigen Läsionsorten und ähnlicher Symptomatik. Allerdings sind zwei Schlaganfall-Läsionen praktisch nie identisch und ein Schlaganfall geht selten mit nur einem Symptom einher. Deswegen ist die Erfassung der Veränderung mehrerer neurologischer Systeme („multipler Netzwerke") notwendig.

Das übergeordnete Ziel der vorliegenden Dissertation war es daher, einen methodischen Ansatz zu entwickeln, der die Plastizität nach einem Schlaganfall auf der Multi-Netzwerk Ebene mittels rs-fMRT beschreiben kann. Dies ermöglicht die Einbeziehung von Patienten mit heterogenen Läsionen und verschiedener klinischer Symptomatik und bietet einen einheitlichen Ansatz für alle Schlaganfall-Patienten, der es dennoch erlaubt, individuelle Verläufe zu identifizieren.

In Studie I wird eine neue mathematische Analyse für rs-fMRT Daten präsentiert, die den sogenannten „Konkordanz-Korrelationskoeffizienten" ermittelt. Dieses Maß beschreibt die räumliche Ähnlichkeit von zwei Konnektivitäts-Kartierungen und erlaubt es somit, Änderungen der Konnektivität im Zeitverlauf zu erkennen. Diese Methode soll für longitudinale Schlaganfall-Untersuchungen anwendbar sein, um große Veränderungen in der funktionellen Konnektivitätsstruktur über die Zeit aufzudecken. Unsere Ergebnisse mit gesunden Kontrollprobanden und frühere Studien, die eine Verbindung zwischen funktionellen

Konnektivitätsveränderung und Verhaltensbeeinträchtigung zeigten, legen nahe, dass Konkordanz ein aussagekräftiges, quantitatives Maß für Plastizität darstellt, wobei sich die Plastizität hier durch Änderungen des räumlichen Musters der Konnektivitäts-Kartierungen ausdrückt.

Basierend auf der methodischen Entwicklung in Studie I wird in Studie II ein Model zur Multi-Netzwerk Analyse der funktionellen Konnektivität nach Schlaganfall präsentiert und auf eine Gruppe von Schlaganfall-Patienten mit heterogenen Läsionen angewandt. Wir berechneten hierzu die Konkordanz auf der Ebene von neuronalen Netzwerken, um Veränderungen in der funktionellen Konnektivität über die Zeit hinweg zu quantifizieren. Wir konnten individuell für die verschiedenen Patienten zeigen – selbst wenn mehrere Netzwerke betroffen waren – dass die Konkordanz sich bevorzugt in von Läsionen betroffenen Netzwerken verringerte. Besonders hervorzuheben ist dabei, dass der Grad der Veränderung der funktionellen Konnektivität („Plastizität") über die Zeit hinweg mit dem Verlauf der klinischen Symptomatik korrelierte. Diese Studie unterstützt das theoretische Model der *Diaschisis* und verallgemeinert die Ergebnisse, die in einzelnen Netzwerken gewonnen wurden, zusätzlich für großräumige Netzwerke; ungeachtet des Läsionsortes oder der einbezogenen Netzwerke.

In Studie III, einer Übersichtsarbeit, werden zunächst die aktuellen Ergebnisse im Feld der rs-fMRT innerhalb der Schlaganfallforschung kritisch diskutiert und evaluiert. Aus dieser kritischen Erörterung leiten wir Perspektiven und Grenzen der Methode ab. Speziell untersuchen wir die mögliche Anwendung von rs-fMRT zur Akutdiagnostik des Schlaganfalls sowie zur Erforschung der Reorganisation des Gehirns in der subakuten und chronischen Phase. Schließlich etablieren wir ein Modell des Schlaganfalls als „Netzwerk-Störung" und erörtern verschiedene Ansätze, welche dieser Sichtweise in der Untersuchung des Schlaganfalls gerecht werden können.

Zusammenfassend schlagen wir einen neuen konzeptuellen und methodischen Ansatz vor, Schlaganfall weniger als lokalisiertes Phänomen sondern vielmehr als Netzwerk-Störung zu verstehen. Diese Sichtweise hat nicht nur wissenschaftliche Implikationen sondern bietet auch therapeutische Perspektiven: Aus Kartierungen der funktionellen Konnektivität nach Schlaganfall können individuell angepasste Protokolle für die nicht-invasive Hirnstimulation abgeleitet werden, mit dem Ziel, Störungen der Netzwerk-Konnektivität zu korrigieren.

List of original research articles

Study I:

Lohmann, G., Ovadia-Caro, S., Jungehulsing, G. J., Margulies, D. S., Villringer, A., & Turner, R. (2012). Connectivity concordance mapping: a new tool for model-free analysis of FMRI data of the human brain. *Frontiers in systems neuroscience, 6*, 13. doi: 10.3389/fnsys.2012.00013.

Study II:

Ovadia-Caro, S., Villringer, K., Fiebach, J., Jungehulsing, G. J., van der Meer, E., Margulies, D. S., & Villringer, A. (2013). Longitudinal effects of lesions on functional networks after stroke. *Journal of cerebral blood flow and metabolism*. doi: 10.1038/jcbfm.2013.80.

Study III:

Ovadia-Caro, S., Margulies, D. S., & Villringer, A. (2014). The value of resting-state functional magnetic resonance imaging in stroke. *Stroke, 45*(9), 2818-2824.doi:10.1161/STROKEAHA.114.003689.

List of abbreviations

fMRI	functional magnetic resonance imaging
rs-fMRI	resting-state fMRI
TMS	transcranial magnetic stimulation
BOLD	blood-oxygen-level-dependent
RSNs	resting-state networks
CCM	connectivity concordance mapping
EEG	electroencephalography
MEG	magnetoencephalography
DTI	diffusion tensor imaging
ROI	region-of-interest
ICA	independent component analysis
DR	dual regression
DWI	diffusion-weighted images
GLM	General linear model
FLAIR	fluid attenuated inversion recovery
NIHSS	national institute of stroke scale
tDCS	transcranial direct current stimulation

List of figures

1 Introduction

The ability of the brain to modify its structural and functional properties is termed *plasticity* (Kolb & Gibb, 2014; Kolb & Whishaw, 1998). This fundamental ability of the brain is at the core of developmental processes, learning, as well as states of disease, among them recovery from brain injuries and stroke (Chen et al., 2002; Cramer et al., 2011; Johansson, 2004; Johnston, 2004; Kolb & Gibb, 2014; Kolb & Whishaw, 1998; Pascual-Leone et al., 2005). The sudden deprivation of blood supply following stroke causes immediate structural damage and behavioral deficits such as hemiparesis, neglect or language deficits. However, the remarkable ability of the brain to change following stroke is at the basis of recovery processes and behavioral improvement. This process is mediated by numerous mechanisms of plasticity (Caleo, 2015; Dimyan & Cohen, 2011; Ko & Yoon, 2013; T. H. Murphy & Corbett, 2009; Nudo, 2013; Overman & Carmichael, 2014).

Areas connected to the stroke lesion play an important role in reorganization after stroke. Outside the localized damage, the structurally intact areas connected to the lesion undergo plastic changes (Carmichael, 2003, 2006; Napieralski et al., 1996; Redecker et al., 2000; Urban et al., 2012). This can provide an explanation for recovery processes as well as explain the complex clinical symptoms that arise and are not solely explained by the local damage. In addition, it can explain why lesions in different locations lead to similar functional deficits, as in the case of neglect (Corbetta & Shulman, 2011). Stroke hence affects a network of interconnected regions rather than a single, localized node (Carter, Shulman, et al., 2012; Corbetta, 2012; Ward, 2005). This view of stroke as a *network disruption* has modified the way in which researchers investigate plasticity following stroke, and has the potential to influence rehabilitation therapy in the future (Alonso-Alonso et al., 2007; Di Pino et al., 2014; Grefkes & Fink, 2011; Grefkes et al., 2010).

Studying multiple networks has become possible with the development of functional magnetic resonance imaging (fMRI), and in particular the study of intrinsic fMRI fluctuations, acquired in the absence of task, known as 'resting-state' fMRI (rs-fMRI). Using rs-fMRI data, connectivity changes can be

characterized at the whole-brain level and the potential role of regions connected to the lesion in the recovery process can be further explored. rs-fMRI has been successfully applied to study connectivity changes in various diseases (Buckner et al., 2009; M. Greicius, 2008; M. D. Greicius et al., 2007; Kelly et al., 2007; Kennedy & Courchesne, 2008; K. Wang et al., 2007), even in severely affected patients suffering disorders of consciousness (Boly et al., 2012; Ovadia-Caro et al., 2012). The technique demands relatively little from the patients during the scan, making rs-fMRI an ideal technique to study connectivity changes after stroke, even in the very acute phase (Amemiya et al., 2013; Lv et al., 2013).

One of the major challenges of exploring plasticity in stroke patients is the heterogeneity of lesions. It is rarely the case that two different patients will present the exact same set of symptoms, or will have the exact lesion location. The research approach until now has been to group patients according to similarity of symptoms (for example, hemiparesis), or similarity of lesion location. This has limited the investigation of plasticity to single networks and single functional domains. The need for a methodology that can be applied to stroke patients with different locations of lesions and different sets of symptoms is evident. Such a method would provide a unified model for exploring plasticity following stroke and has the potential for better characterizing symptoms and providing a basis for guided therapy in the form of non-invasive brain stimulation techniques.

In this dissertation, I will review the three studies conducted during my doctoral research that address the topic of plasticity following stroke, and in particular suggest a generalized methodological framework to study recovery in patients with heterogeneous lesions. Study I presents a data-driven mathematical tool to analyze rs-fMRI data. This algorithm quantifies the reproducibility of connectivity patterns using the concordance correlation coefficient. The feasibility and potential usage of this novel analysis is demonstrated on a group of healthy controls, as well as on a longitudinal dataset of a stroke patient (Lohmann et al., 2012). Study II focuses on the longitudinal recovery of functional networks following ischemic stroke, and the relationship between signal alterations and behavioral recovery. Based on the suggested concordance measure, we developed a methodological approach that explores connectivity

changes in multiple networks over time, taking into account heterogeneous lesions affecting more than one network (Ovadia-Caro et al., 2013). Study III is a review paper exploring the potential usefulness and limitations of the application of rs-fMRI data to study stroke patients. Being a relatively new field, all studies published thus far are reviewed and the main results are summarized and discussed. The general methodological trend in the field and in the understanding of stroke as a network disruption is described and the clinical utility of rs-fMRI in patients with stroke is discussed in detail (Ovadia-Caro et al., 2014).

2 Theoretical background

2.1 General overview of stroke

Stroke is defined as a neurological deficit caused by a sudden localized injury to the central nervous system due to a vascular pathology (Sacco et al., 2013). According to the updated report from the American Heart Association, stroke was the second-leading cause of death in 2010, accounting for 11.13% of total deaths worldwide. Currently, stroke is the fifth highest cause of death in the United States, with a killing rate of once every four minutes. Importantly, stroke is the main cause of long-term disability, and the leading preventable cause of disability (Mozaffarian et al., 2015).

There are two main types of stroke, *ischemic stroke* is caused by obstruction of a blood vessel and is the most prevalent type of stroke (87% of all strokes), and *hemorrhagic stroke* occurs due to blood vessel rupture (Mozaffarian, et al., 2015). The data presented in this dissertation is of patients following ischemic strokes.

Immediately following stroke, a complex cascade of pathophysiological changes occurs in the area of infarct and in the surrounding tissue. Excitotoxicity and peri-infarct depolarization are followed by inflammation and apoptosis. In parallel, various endogenous attempts to protect the tissue are initiated (Dirnagl, 2012; Dirnagl & Endres, 2014; Dirnagl et al., 1999; Endres et al., 2008; Endres et al., 2004; Kunz et al., 2010). Recovery of function at this stage is dependent on reperfusion, as well as on the complex interaction between adaptive and maladaptive changes taking place as part of the pathophysiological cascade.

Following the acute phase, functional recovery is attributed to reorganization and plastic processes in functioning areas. Since reorganization also occurs in remote areas connected to the lesion (Carter, Shulman, et al., 2012; Corbetta, 2012; Ward, 2005), therapeutic attempts such as non-invasive brain stimulation are aimed at modulating the connectivity of affected brain networks with the aim of enhancing reorganization in structurally intact areas (Alonso-Alonso, et al., 2007; Di Pino, et al., 2014; Grefkes & Fink, 2011; Grefkes, et al., 2010; Sehm et al., 2012).

2.2 Theories of plasticity following stroke

2.2.1 Understanding stroke: from localization to network disruption

The traditional clinical view to the outcome following stroke is attributing the functional damage to the locally affected area. This is well supported by early lesion studies going back to Paul Broca's description in the 1860s of language production impairment (i.e. aphasia) following lesion in the left frontal lobe (Broca, 1861b; Dronkers et al., 2007). Lesion studies have provided extensive knowledge concerning localization of function and specialization of various areas in the brain, laying the foundations for modern neuropsychology and cognitive neuroscience (Dronkers, et al., 2007). However, localization of functions disregards a very important part of stroke pathology; the contribution of network-wide changes (Carter, Shulman, et al., 2012).

Some evidence suggests that the local damage cannot fully explain the full-blown complex clinical symptoms after stroke, and most importantly does not explain the recovery from clinical symptoms. Additionally, similar symptoms can arise from different locations of lesions (Corbetta & Shulman, 2011). This discrepancy may be partially explained by distal effects of the localized lesion on areas connected to it (Feeney & Baron, 1986; He, Shulman, et al., 2007; Rossini et al., 2003). These findings, along with a general view of normal brain function as an interaction between different interconnected brain regions has initiated a shift in the way stroke is understood and investigated (Carter, Shulman, et al., 2012; Corbetta, 2012; Ovadia-Caro, et al., 2014; Ward, 2005).

Stroke is currently considered as a *network disruption* rather than a mere local, single-node lesion. *Network disruption* (see Figure 1 for schematic illustration) is reflected in alterations of connections within the affected network, even between structurally intact regions, leading to the complex functional deficits. Connections between the lesion sites and their interconnected regions are permanently broken. In addition, connections between structurally intact regions that are part of the affected network are temporarily impaired, and the recovery of these connections is correlated with behavioral improvement (He, Snyder, et al., 2007). Reorganization can be more generally characterized at the network-level in order to better understand and enhance recovery.

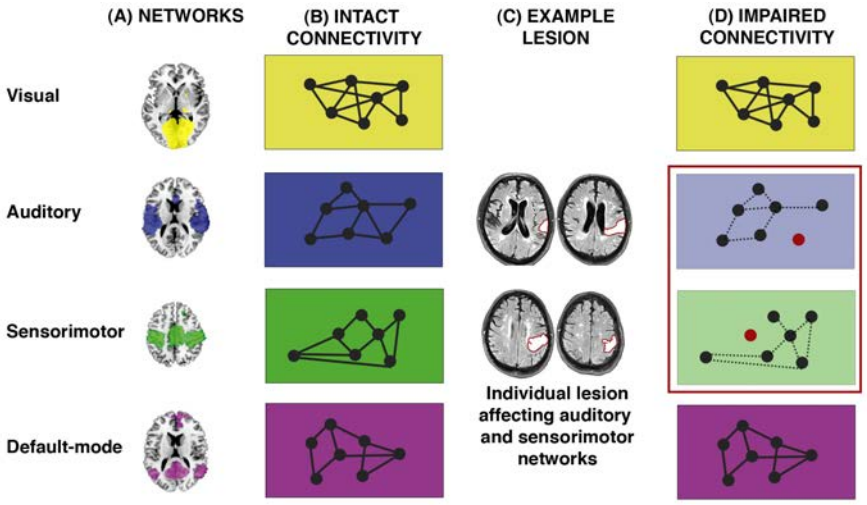

Figure 1. **Schematic illustration of network disruption after stroke.**
(A) Functional networks are correlated even in the absence of task. Here, an example of 4 functional networks based on resting-state functional MRI functional connectivity in healthy controls. (B) Intact connectivity structure in healthy controls is reflected in the high correlation (solid black lines) between functionally relevant nodes of a specific network. (C) Anatomic location of an individual lesion in a patient with a recent ischemic stroke (white areas outlined in red). The lesion affects the auditory and the sensorimotor networks, sparing the visual and default-mode networks. (D) After stroke, structural damage to specific nodes (red circles) in the network leads to global disruption in connectivity in the affected networks (red rectangle), even in structurally intact regions. Connectivity is interrupted from the lesion area and altered among the structurally intact nodes of the network (black dotted lines). The disruption of connectivity after a local stroke is network-specific and largely spares the unaffected functional networks. A multi-network assessment of changes in functional connectivity has better potential for reflecting complex clinical symptoms, which often involve more than one functional network. Figure adapted from study III (Ovadia-Caro, et al., 2014).

2.2.2 Diaschisis: Connectivity as a mediating mechanism for network disruption and reorganization

The idea that localized lesions have distal effects on structurally intact interconnected areas is not new. Von Monako (1914) described in the 19th century the phenomenon of *diaschisis,* in which, following stroke and other types of acute brain injuries, regions that are far from the lesion yet connected to it demonstrate neural depression or what he called 'functional stand-still' (Andrews, 1991; Finger et al., 2004; von Monakow, 1914, 1969). The theory was

initially described to explain symptoms after stroke that were not easily attributed to the localized lesion, as well as to explain recovery from what he referred to as 'transient symptoms'.

It was only later on that supporting findings for metabolic diaschisis were additionally described (Finger, et al., 2004). Nowadays, the term is usually used to describe both neuronal and metabolic alterations in distant areas connected to the lesion (Andrews, 1991; Finger, et al., 2004). While some aspects of the theory of diaschisis as it was initially described are debatable (Andrews, 1991; Dobkin et al., 1989; Finger, et al., 2004; Markowitsch & Pritzel, 1978; West, 1978; West et al., 1976), its main relevance today is in drawing attention to the structurally intact regions connected to the lesion as being a central aspect of the pathology, as well as suggesting that connectivity plays a crucial role in symptoms manifestation and recovery processes (Carrera & Tononi, 2014).

2.3 Empirical evidence for diaschisis and network disruption

2.3.1 Plasticity at the cellular level following stroke

Evidence from animal studies suggests that at the cellular level, axonal sprouting and migration of neuroblasts from the subventricular zone are the two main regenerative events taking place within the periinfarct region (i.e. the area surrounding the lesion) (Carmichael, 2006; Carmichael, Saper, et al., 2016; Carmichael et al., 2001; Dancause et al., 2005; Napieralski, et al., 1996). Other than these localized changes, axonal sprouting has been reported to take place in remote areas connected to the lesion in particular in the homotopic contralateral region (Carmichael, 2003, 2006, 2008; Carmichael, Kathirvelu, et al., 2016; Carmichael, et al., 2001; Dancause, et al., 2005; Napieralski, et al., 1996). Moreover, Carmichael and colleagues have found that axonal sprouting after ischemia is induced by intrinsic patterns of synchronous low-frequency neuronal activity in areas connected to the infarct core (Carmichael & Chesselet, 2002). These findings provide a cellular-based rationale for exploring connectivity changes following stroke within affected yet intact areas using other modalities,

such as rs-fMRI, and suggest an underlying neurophysiological mechanism for connectivity changes following ischemia.

2.3.2 fMRI studies of plasticity following stroke

While detailed animal work as presented above can provide potential pathophysiological mechanisms, fMRI is used to investigate plasticity in vivo at the macroscale (Sporns et al., 2005).

Task-based fMRI paradigms, or activation studies, have been employed to investigate activation patterns following stroke in response to a specific task (Corbetta et al., 2005; Saur & Hartwigsen, 2012; Saur et al., 2006; Ward & Cohen, 2004). This has mainly been done using motor tasks, for example moving the paretic hand (Grefkes et al., 2008; Tombari et al., 2004; Ward et al., 2003), but also language (Kiran, 2012; Saur, et al., 2006; Saur et al., 2010) and visuospatial tasks (Corbetta, et al., 2005). Abnormal levels of cortical activation have been reported for patients after stroke in the affected hemisphere as well as in the contralateral homologues, both in the subacute and the chronic phase (Corbetta, et al., 2005; Grefkes & Fink, 2011; Saur & Hartwigsen, 2012; Saur, et al., 2006). Up-regulation or down-regulation of activation levels in both contralateral homologues and peri-lesion areas has been suggested to be related to the time that has passed since stroke onset, possibly reflecting different stages in the dynamics of recovery (Grefkes & Ward, 2014; Saur, et al., 2006) as well as to the degree of motor impairment (Rehme et al., 2011).

Up-regulation of activity in the contralesional hemisphere (Corbetta, et al., 2005; Rehme et al., 2012; Saur, et al., 2006) forms the basis for using stimulation techniques such as transcranial magnetic stimulation (TMS) to regain 'balanced' activation levels in affected and unaffected hemispheres (Grefkes & Fink, 2011, 2012; Grefkes & Ward, 2014; Hummel & Cohen, 2006; Meinzer et al., 2016). However, results are mixed in terms of the effectiveness of such treatment, leading in some cases to the deterioration of symptoms (Ameli et al., 2009; Grefkes & Ward, 2014).

Activation studies can provide information concerning localization of a specific function, but they do not provide information concerning the process of

integration and the interplay between the different regions involved, which is an inseparable part of the function (K. J. Friston, 1994; Raichle, 2009). Connectivity analysis based on rs-fMRI can inform us about temporal synchronization between different regions that are part of the same network (Sporns, et al., 2005) and is hence a complementary approach to task-based fMRI.

A central caveat to task-based paradigms is performance bias, an issue of high relevance to patient populations in general and stroke patients in particular. The fact that levels of activation are different between patients and controls may be driven by the inability of patients to adequately perform the task at hand, leading to artifactually 'decreased' activation (Carter, Shulman, et al., 2012). Additionally, using task-based paradigms one cannot examine severely affected patients, as they might be unable to perform the task. A task-free approach such as rs-fMRI eliminates performance bias and provides a more controlled experimental approach to study stroke patients, even in those severely affected.

2.3.3 rs-fMRI studies of plasticity following stroke

rs-fMRI is a task-independent neuroimaging method based on the recording of slow (typically <0.1Hz) on-going intrinsic oscillations in the blood oxygen-level dependent (BOLD) signal (M. D. Fox & Raichle, 2007; Raichle, 2009). This signal can be used to compute *functional connectivity,* defined as the temporal correlation of the BOLD signal from different brain regions (B. Biswal et al., 1995; Lowe et al., 1998). Functional connectivity hence yields a spatial map representing the statistical interdependence between regions (K. J. Friston, 1994, 2011). Functional connectivity based on rs-fMRI has been shown to be a highly reliable and reproducible measure, even on the individual level (B. B. Biswal et al., 2010; J.S. Damoiseaux et al., 2006; Shehzad et al., 2009).

Even in the absence of task, functional connectivity is increased between areas that are part of the same network and are jointly activated during specific tasks (B. Biswal, et al., 1995). These emerging networks are also referred to as resting-state networks (RSNs) (Smith et al., 2009).

The main advantage of rs-fMRI is the ability to explore multiple RSNs post hoc using various connectivity analyses (Margulies et al., 2010). Additionally, the

relative simplicity of data collection and minimal demands from the participants makes rs-fMRI an ideal non-invasive experimental tool for investigating various clinical populations (M. D. Fox & Greicius, 2010).

Following stroke, rs-fMRI has been successfully applied to study plasticity and reorganization using functional connectivity. The main conclusion arising from animal studies (van Meer et al., 2012; van Meer, van der Marel, Otte, et al., 2010; van Meer, van der Marel, Wang, et al., 2010), modeling work (Alstott et al., 2009; Honey & Sporns, 2008) and human studies (Carter et al., 2010; Carter, Patel, et al., 2012; Golestani et al., 2013; He, Snyder, et al., 2007; Nomura et al., 2010; Park et al., 2011; Tuladhar et al., 2013; L. Wang et al., 2010) is that functional connectivity is reduced in structurally intact areas connected to the local lesion. This phenomenon has been shown in the motor, sensory, attention and default-mode networks. This breakdown of connectivity in intact yet connected areas supports the theory of diaschisis and the current views of stroke as a network disruption.

Most importantly, changes in functional connectivity following stroke correlate with behavioral symptoms in many of these studies. He and colleagues have shown that in stroke patients with symptoms of neglect functional connectivity is impaired in structurally intact areas that are part of the attention network. They additionally found that when symptoms of neglect recovered, functional connectivity between the observed regions increased (He, Snyder, et al., 2007). Such findings further strengthen the functional significance of rs-fMRI connectivity changes after stroke and their potential therapeutic and prognostic value.

2.3.4 From single network to multiple networks – the challenge of heterogeneity

Animal work and human studies conducted thus far using rs-fMRI explored changes in functional connectivity in a single functional domain, mainly in the motor, using a relatively small number of regions for the analysis. Most studies explored changes in inter-hemispheric connectivity. While connectivity between homologous regions is indeed a very robust and consistent finding in rs-fMRI

connectivity data (Stark et al., 2008), such analysis makes use of very small parts of the connectivity matrix available in the data and enables us to explore only a single network.

One of the reasons the field has been focused on single network analysis is the attempt to overcome the challenge of lesion heterogeneity. In most studies, patients are grouped based on the similarity of their lesion locations or clinical symptoms (for example, paresis). However, it is rarely the case that two structural lesions will be identical and stroke typically affects more than one functional network and hence, more than one functional domain. A more generalizable global model is needed to study plasticity in stroke patients in order to create a more realistic picture of the involved networks. Such a global model that is based on a whole-brain analysis would also maximize the utilization of the raw connectivity data (Sporns, 2011; Sporns, et al., 2005). Importantly, a unified model that can take into account different locations of lesions and multiple-network damage may provide a multifaceted basis for individualized therapy in the form of stimulation techniques.

3 Research questions and hypotheses

This thesis aims to investigate plasticity following stroke using rs-fMRI functional connectivity at the whole-brain level. Since the majority of studies conducted in the field thus far have addressed connectivity changes following stroke in a single network, and in most cases using few regions-of-interest, the need for an approach that can be applied to heterogeneous lesions affecting more than one network is evident.

Study I presents a novel data-driven algorithm to analyze rs-fMRI data. We introduce the quantification of consistency (reproducibility) of functional connectivity patterns at the whole-brain voxel level using the concordance correlation coefficient. In this study, we describe in detail our suggested algorithm, termed connectivity concordance mapping (CCM). We apply CCM in two different experimental settings to demonstrate its applicability and functional significance. CCM can be applied both in an inter-subject and intra-subject manner and can be used as a data-driven approach for guiding second level analyses.

Specifically, in the first experiment CCM was computed for rs-fMRI data collected using eyes closed and eyes open conditions in healthy controls with the aim of providing a proof of concept for the algorithm's use and demonstrating the functional relevance of concordance. Based on previous findings we expected:

1. A decrease in concordance within visual areas when CCM is applied to the two experimental conditions in an intra-subject manner (McAvoy et al., 2008; Van Dijk et al., 2010).
2. When CCM is applied in an inter-subject manner, areas of larger anatomical variability (such as frontal regions) are expected to show decreased concordance (Fischl et al., 2008; Mueller et al., 2013).
3. Areas of spurious signals (such as the white matter) are expected to demonstrate decreased concordance compared to grey matter due to non-neuronal signal in these areas expressed by higher levels of random noise (Weissenbacher et al., 2009).

In the second experiment CCM was applied to rs-fMRI longitudinal dataset from a single stroke patient. Here, the algorithm was used for a data-driven analysis

in order to detect regions demonstrating a large change in the functional connectivity pattern over time. Although this experiment had no prior hypothesis due to the data-driven nature of the algorithm, the findings are discussed in the context of previous literature and our theoretical model of plasticity following stroke. The results based on CCM were used for a second-level seed-based connectivity analysis to further explore the dynamic changes in connectivity over time.

Study II explores the longitudinal connectivity changes induced by lesions and their relationship to functional recovery. Based on the concordance method developed in study I, we present an approach that can be applied to a group of stroke patients with varying locations of lesions affecting more than one functional network. We explored changes in functional connectivity over time in eight representative functional networks. Additionally, we explored the relationship between changes in connectivity over time and the trajectory and degree of functional recovery.

Taking into account the theoretical model of diaschisis (Andrews, 1991; Finger, et al., 2004), computational models (Alstott, et al., 2009; Honey & Sporns, 2008) as well as previous empirical findings obtained in single networks (Carter, et al., 2010; Carter, Patel, et al., 2012; Golestani, et al., 2013; He, Snyder, et al., 2007; Park, et al., 2011; L. Wang, et al., 2010), we hypothesized that:

1. Lesions will have a more detrimental effect on functional connectivity in intact areas connected to the lesion (i.e. affected networks) as compared to more loosely connected areas (i.e. unaffected networks).

In addition, it has been previously shown that functional connectivity changes in individual networks are correlated with behavioral impairments in specific tasks (Carter, et al., 2010; Golestani, et al., 2013; He, Snyder, et al., 2007; Park, et al., 2011; van Meer, et al., 2012; van Meer, van der Marel, Wang, et al., 2010; L. Wang, et al., 2010). We hence hypothesized that:

2. The degree of changes in connectivity observed over time in multiple affected networks will be positively correlated with the degree of behavioral change over time.

The application of rs-fMRI to study plasticity following stroke is a relatively new field. **Study III** is a review paper aimed at investigating the usage of rs-fMRI to study stroke in general, and plasticity following stroke in particular. The potential clinical value, main findings and limitations are described and discussed in detail. The view of stroke as a network disruption suggested by our empirical findings is discussed in the context of previous studies and new trends and directions for future research are suggested. In addition to reviewing and summarizing the current literature, this work suggests a framework for exploring plasticity in patients following stroke in the aim of promoting the understanding of recovery processes.

4 General methodological approach

4.1 The restless brain: intrinsic brain activity

The brain is constantly active. Even in the absence of explicit sensory tasks, neurons are spontaneously firing (Arieli et al., 1995; Arieli et al., 1996; Grinvald et al., 2003; Kenet et al., 2003; Leopold et al., 2003; Llinas, 1988; Miller et al., 2014), and the brain's consumption of energy is very high (Raichle, 2006, 2015; Raichle & Mintun, 2006). Although early electrophysiological recordings by Hans Berger linked intrinsic brain activity to various cognitive states such as sleep, wakeful rest and alertness (Berger, 1929, 1969), most studies following that period were focused on the evoked activity induced by controlled sensory stimuli. In these experiments intrinsic brain activity was regarded as mere noise, as the evoked response was based on averaging activity over multiple repetitions. The brain in these experiments was treated as a reflexive organ producing a deterministic output in response to an environmental input (Burke, 2007).

Interestingly, the local change in blood flow induced by evoked activity is surprisingly small, often less than 5% from baseline activity (Raichle & Mintun, 2006). A clear question that follows is why the brain is in need of such high levels of energy to maintain intrinsic activity and the precise cognitive role, if any, of this spontaneous activity. Supportive of the proactive view of the brain, some authors suggest that intrinsic activity plays an important role in prediction and prior representations embedded in the brain (Bar, 2007, 2009; Engel et al., 2001; K. Friston & Kiebel, 2009; Yuste et al., 2005). Evidence for this can be found in animal studies suggesting intrinsic activity can account for inter-trial variability and prior representations (Arieli, et al., 1995; Arieli, et al., 1996; Grinvald, et al., 2003; Kenet, et al., 2003; Leopold, et al., 2003; Llinas, 1988; Miller, et al., 2014). Similar results have been reported in human studies, where the intrinsic activity prior to stimuli appearance had an influence on the perception of the stimuli (Becker et al., 2011; M. D. Fox et al., 2007; M. D. Fox et al., 2006; G. Hesselmann et al., 2008; G. Hesselmann, Kell, C.A., Eger, E., Kleinschmidt, A., 2008). In contrary to this possible cognitive role, high levels of intrinsic activity have been recorded

during non-REM sleep and under anesthesia (M. D. Greicius et al., 2008; Nir et al., 2008; Vincent et al., 2007).

Although the exact role of intrinsic activity is still under debate (Raichle, 2015), in the last decade it has been repeatedly shown that intrinsic fluctuation are of clinical value for both diagnosing and exploring various states of disease (M. D. Fox & Greicius, 2010; Gillebert & Mantini, 2013).

4.2 Resting-state fMRI: measuring intrinsic activity using the BOLD signal

Intrinsic activity can be measured in humans using non-invasive techniques. Using fMRI, intrinsic activity is measured as task-independent BOLD signal fluctuations (M. D. Fox & Raichle, 2007). This on-going recorded BOLD signal in the absence of explicit task is commonly referred to as resting-state fMRI (rs-fMRI) signal. The term resting-state to describe on-going BOLD fluctuations is widely used in the field due to the absence of experimental task and has therefore been used throughout this dissertation as well. However, one should note that this terminology is somewhat misleading, as these fluctuations are robust and occur in all areas of the cortex at amplitudes similar to task-evoked activity (Nir et al., 2006), suggesting this is not a resting-state per se, rather a state in which no explicit task is imposed on the subject.

rs-fMRI has specific and reliable characteristics (Shehzad, et al., 2009). In the temporal domain intrinsic fluctuations as measured by the BOLD contrast exhibit slow (usually between 0.01-0.1Hz) fluctuations that are not simply an artifact of physiological activity, breathing, or vasculature (Birn et al., 2006). In the spatial domain, rs-fMRI is not random, but rather organized such that areas that are jointly active during task are correlated in their intrinsic activity, even in the absence of task demands (B. Biswal, et al., 1995). The term to describe this statistical interdependence, or synchrony, between distant regions is *functional connectivity* (K. J. Friston, 1994). Functional connectivity is computed as the temporal correlation between intrinsic BOLD signal from different regions of interest.

In 1995, Biswal and colleagues were the first to explore functional connectivity in the motor network in the absence of task demands using rs-fMRI. The spatial pattern emerging from the functional connectivity analysis in the motor network replicated the familiar spatial pattern of activation during a finger-tapping task (B. Biswal, et al., 1995). This seminal work has sparked a large field of research (Birn, 2012; Raichle, 2015) and was the first to draw attention to the fact that intrinsic BOLD signal contains meaningful information. Similar to Biswal's finding in the motor network, other areas that are part of specific functional domains during tasks have been shown to correlate in their intrinsic activity in the absence of task demands. These include visual, sensorimotor, auditory, language and default-mode networks and are commonly referred to as resting-state networks (RSNs) (Beckmann et al., 2005; Smith, et al., 2009). RSNs have been shown to be highly reproducible across individuals, time and scanning sites (B. B. Biswal, et al., 2010; J.S. Damoiseaux, et al., 2006; Shehzad, et al., 2009).

Recent electrophysiological studies using electroencephalography (EEG) and magnetoencephalography (MEG) in humans have provided support for an underlying neural basis of connectivity measured by rs-fMRI. Simultaneous EEG-fMRI recordings have demonstrated that intrinsic BOLD signal can be predicted by the power in the high frequencies of electrical activity (Laufs & et al., 2003; Laufs et al., 2006; Laufs et al., 2003; Moosmann et al., 2003). Mantini and colleagues have further described specific electrophysiological signatures for individual, well-known RSN networks (Mantini et al., 2007). Similarly, MEG studies have reported correlation between large-scale cortical networks that are spatially similar to RSNs and power mainly in the alpha and beta frequencies (Brookes et al., 2011; de Pasquale et al., 2010; Hipp et al., 2012). Taken together, these studies further support the importance of synchronous activity for communication between neurons across distant spatial locations (Engel, et al., 2001).

The functional significance of RSNs is additionally supported by the strong link between functional connectivity and the underlying structural connectivity measured in humans using diffusion tensor imaging (DTI). Multiple studies have explored the link between these two measures and reported positive correlation (J. S. Damoiseaux & Greicius, 2009; M. D. Greicius et al., 2009; Hagmann et al.,

2008; Honey et al., 2009; Horn et al., 2014; Skudlarski et al., 2008; M. van den Heuvel et al., 2008; M. P. van den Heuvel et al., 2009). While a strong link between these two measures is evident, it appears that direct/monosynaptic structural connectivity cannot fully account for observed functional connectivity maps. Since functional connectivity is based on measures of temporal correlation, a high correlation can reflect indirect connections via a third region (M. D. Greicius, et al., 2009; Hagmann, et al., 2008; Honey, et al., 2009; Vincent, et al., 2007). In addition, functional connections are not stationary while the anatomical structure is (Honey et al., 2007; Hutchison, Womelsdorf, Allen, et al., 2013). Functional connections between different brain regions are hence constrained by the underlying anatomical connections, yet cannot be fully accounted for based on anatomy alone (Honey, et al., 2009).

4.3 Analysis of functional connectivity: methodological tools

Functional connectivity using rs-fMRI aims at describing the relationships between inter-connected regions. To date, several methodological tools are in use to compute functional connectivity. With the development of rs-fMRI field and the general development in computational abilities, methods to analyze rs-fMRI data have evolved (for a detailed review see (Margulies, et al., 2010)). In this section, I will briefly discuss the methods used throughout this dissertation including their advantages and potential limitations.

The most basic analysis, and perhaps the most commonly used approach is termed seed-based, or region-of-interest (ROI) analysis. This analysis entails computing an average BOLD signal over a priori defined areas and then computing the correlation (usually Pearson's correlation coefficient) between them to depict the strength of connections. Biswal and colleagues performed such an analysis in the initial study from 1995 (B. Biswal, et al., 1995). The main advantage of seed-based connectivity is that this analysis is relatively simple and hence drawing conclusions from it is straightforward. However, this analysis requires an a priori decision on the areas of interest and is usually then limited to a relatively small number of regions. It should therefore be guided by either a strong hypotheses based on existing literature or by an initial data-driven

approach to detect areas relevant for further exploration (Margulies, et al., 2010). We have used this approach in study I based on results from our data-driven algorithm (see section 5.1.3, experiment B).

Contrary to the seed-based approach, independent component analysis (ICA) is a data-driven approach. ICA is based on blind source separation or decomposition techniques. The data is delineated into maximally independent components in the spatial domain. Each component has a time course and a corresponding spatial map (Beckmann, et al., 2005; Kiviniemi et al., 2003; van de Ven et al., 2004). Since ICA is a data-driven approach, it does not require an a priori hypothesis and is hence suited for exploratory analysis. In addition, unlike seed-based analysis, it does not require any preprocessing of the data. However, it does require a posteriori selection of functionally meaningful components over noise components. This decision is usually done by visual inspection. In addition, the number of components that results from the ICA is pre-defined by the user in most cases, and is arbitrary since the "real" number of components in the brain is unknown (Margulies, et al., 2010). Although seed-based analysis and ICA are different analytic approaches, they produce similar RSNs, supporting the meaningfulness and high consistency of these networks (Joel et al., 2011).

Based on ICA, dual regression (DR) has recently been suggested as an approach for more accurate group-level comparison. DR runs a group average ICA and then estimates the individualized version of each of the group-level spatial maps. This approach can also be used with a pre-defined set of networks that were computed on a different population than the population under observation. The analysis is done in two regression steps; first, the group spatial maps are used as regressors on each subject's four-dimensional dataset. This results in a set of time courses. Second, these time courses are used as regressors on the same four-dimensional data set to get a set of individualized, subject-specific spatial maps. The resulting maps can be used for further group-level comparison (Beckmann et al., 2009; Filippini et al., 2009). We have used DR analysis as part of our analysis in study II (see section 5.2.2).

CCM as introduced in study I is an additional, novel data-driven method to explore the reproducibility of functional connectivity patterns at the whole-brain voxel-level. The pairwise correlation matrix is computed at the voxel level and

reproducibility of connectivity patterns is estimated in an inter-subject or intra-subject manner using Kendall's W concordance correlation coefficient. The main novelty of CCM is that it takes the whole connectivity matrix into account (i.e. a matrix of approx. 50,000*50,000 connections), making use of all the data (Lohmann, et al., 2012). Other recent approaches to characterize topological traits of the connectivity matrix (i.e. graph theory approach) require dimension reduction due to their large computational complexity (Bullmore & Sporns, 2009). Reduction of dimensions is usually done using an a priori defined atlas template. While there are both anatomically-based (Tzourio-Mazoyer et al., 2002) and functionally-based (Craddock et al., 2012) atlases, the decision of which atlas should be used for a specific study/research question is not trivial and this decision has a direct impact on the results. On the contrary, performing the analyses at the voxel-level makes no assumptions concerning the 'correct' template to be used and is hence preferable.

4.4 rs-fMRI as a clinical tool to study pathologies and plasticity

The ability to explore multiple RSNs post hoc and the relatively minimal demands from the subjects makes rs-fMRI an ideal non-invasive experimental approach to study various clinical populations. Indeed, alterations in rs-fMRI functional connectivity have been described in various states of disease among them Alzheimer's disease (M. D. Greicius et al., 2004; Rombouts et al., 2005; K. Wang, et al., 2007), autism (Cherkassky et al., 2006; Kennedy & Courchesne, 2008), schizophrenia (Bluhm et al., 2007; Zhou et al., 2007), depression (Anand et al., 2005; M. D. Greicius, et al., 2007), congenital blindness (Striem-Amit et al., 2015), disorders of consciousness (Boly et al., 2012; Ovadia-Caro et al., 2012; Vanhaudenhuyse et al., 2010), as well as stroke (Carter, Shulman, et al., 2012; Gillebert & Mantini, 2013; Ovadia-Caro, et al., 2014). Importantly, changes in rs-fMRI functional connectivity have been shown to correlate with behavioral impairments in many of these clinical populations, including after stroke, strengthening the clinical significance of this tool (M. D. Fox & Greicius, 2010).
The main advantages of rs-fMRI to study plasticity in stroke patients is that unlike task-based fMRI designs, even severely affected patients can be studied.

The task-free nature of rs-fMRI eliminates task-performance bias and patients can be studied even in the acute phase after injury (Amemiya, et al., 2013; Lv, et al., 2013), a point of importance when studying plasticity and recovery processes. The ability to explore in a single scan multiple RSNs is a point highly relevant to study plasticity in stroke patients, since in most cases after stroke, more than one functional domain is impaired and more than a single network is affected. A whole-brain approach can provide a more accurate model of symptoms and can provide a basis for individualized therapy in the form of stimulation techniques (Opitz et al., 2016). rs-fMRI hence provides a promising, clinically valuable approach to study large-scale organization and plasticity induced by stroke (Carter, Shulman, et al., 2012).

5 Summary of related papers

5.1 Study I: Connectivity concordance mapping: a new tool for model-free analysis of fMRI data of the human brain (Lohmann, et al., 2012)

5.1.1 Background

rs-fMRI data can be used to derive information on the relationship between distant, connected brain regions. This analysis of functional connectivity can be computed using various post-processing methodologies (Margulies, et al., 2010). While some approaches require a priori hypothesis concerning the areas of interests (i.e. seed-based analysis), other popular data-driven approaches require a-posteriori decision on distinguishing functionally relevant areas from spurious ones (i.e. ICA). Recent developments in the field analyze rs-fMRI data using graph-based theory computations (Buckner, 2010; Bullmore & Sporns, 2009; Sporns, 2011), but this approach requires dimension reduction due to computational complexity (Craddock, et al., 2012; Tzourio-Mazoyer et al., 2002). An approach that could be computed on the whole-brain voxel level would take the full connectivity matrix into account and maximize the utilization of the raw data. Here, we introduce the CCM approach to analyze rs-fMRI data. The main purpose of CCM is quantifying the reproducibility of whole-brain patterns of connectivity at the single-voxel level. Voxels with reproducible connectivity patterns may be of interest and can be the subject of further analysis. CCM can be applied to both inter- and intra-subject datasets. In this study, the suggested algorithm is described and applied to data from healthy controls as well as a single stroke patient to demonstrate its applicability. Reproducibility of fMRI activity has been previously shown to be of functional relevance (Hasson et al., 2010); however, it has not been applied during resting conditions and has not been computed for each voxel. The main novelty of our suggested approach is that CCM is computed at the voxel level, taking into account the full connectivity matrix of the whole brain.

5.1.2 Methods

Algorithm

The first step of computing CCM is the definition of a mask covering the whole brain. In our study, this mask contained approximately 50,000 voxels (n voxels). All further analysis steps are performed on this mask. As described earlier, CCM is computed between different rs-fMRI datasets. For each dataset, for each single voxel, a vector of pairwise similarity is computed. We used Pearson's linear correlation coefficient as our similarity measure. This step is repeated for each voxel in the mask, yielding n similarity vectors. We next compute for each vector the agreement of the connectivity pattern between different measures. We use Kendall's W as a measure of concordance as it does not assume Gaussianity of the data. Kendall's W produces a value between 0 and 1 such that 1 represents complete agreement and 0 represents complete disagreement. This value of concordance is assigned back to the specific voxel creating a whole-brain voxel-wise map of concordance values determining the reproducibility of spatial connectivity pattern for that particular voxel (see Figure 2 for description of the algorithm flow). It is important to note that concordance is different from correlation such that it does not subtract the mean during computation. This means that the absolute value of pairwise correlation influences the concordance value, while in the case of correlation a similar pattern is the determining factor. For example, in a case where the connectivity pattern is similar but has different underlying correlation values (for example generally lower correlation values), spatial correlation will provide a high value while spatial concordance will be low. This information can of course be of importance especially when comparing patients with healthy controls.

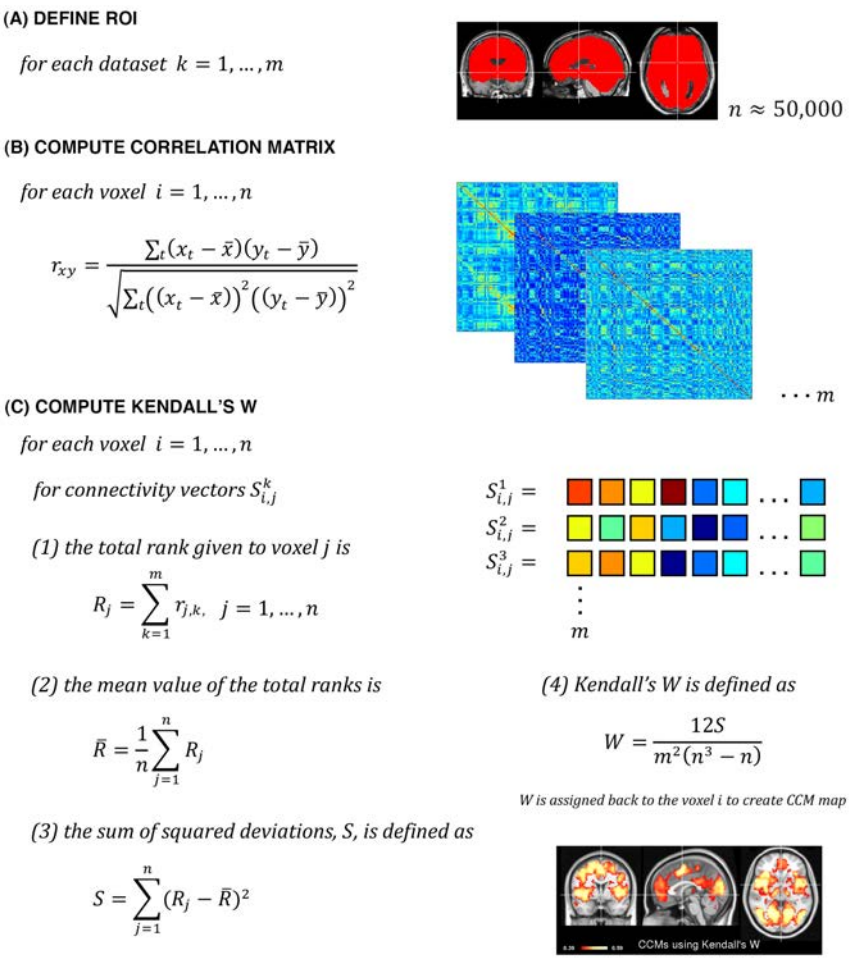

Figure 2: **CCM algorithm.**

(A) Definition of ROI using a whole brain mask. n denotes the number of voxels in the ROI. (B) Computing correlation matrices for each dataset (k) and for each voxel (i). (C) Computing Kendall's W for each voxels' connectivity vector over the multiple measurements using the following steps: ranking, computing mean value of total ranks and the sum of squared deviation prior to the computation of Kendall's W. The value of Kendall's W for each voxels' connectivity pattern over the multiple datasets is assigned back to the voxel to create the CCM map.

Experiment A: applying CCM to healthy controls data

In order to demonstrate the applicability of the CCM algorithm, it was first applied to rs-fMRI data from healthy controls (N=7). Although rs-fMRI does not

involve an explicit task, there are different approaches in the field as to whether subjects' eyes should be open or closed. Here, we applied CCM to rs-fMRI data from both conditions. CCM was computed (1) inter-subject for each condition and (2) intra-subject between different conditions.

Experiment B: Applying CCM to a longitudinal dataset following stroke

In order to explore the possible clinical applicability of CCM and demonstrate the utility of the algorithm as a data-driven method, CCM was applied to longitudinal rs-fMRI data from a single patient following stroke. Data was collected at four time points; day 1, day 27, day 94 and day 199 following stroke. Diffusion-weighted images (DWI) from day 1 were used for lesion localization. The infarct was located in the left thalamus and left occipital cortex. CCM was computed for all time points and also for pairs of consecutive time points. Based on the CCM whole brain results, seed-based analysis was performed as a second-level data guided analysis. The seed area was located in bilateral globus pallidus, where concordance values were lowest (local minima).

5.1.3 Results

Experiment A:

Inter-subject CCM for the healthy control sample detected large proportions of the grey matter as concordant. Areas of spurious signal, such as the white matter, showed low concordance values. This was the case for both conditions (eyes open and eyes closed). Regions that are known to be part of the default-mode network (M. D. Fox et al., 2005; Golland et al., 2007) were less concordant than primary sensory areas. Prefrontal regions, especially in the lateral surface were the least concordant in their connectivity pattern. To explore concordance between the conditions, CCM was computed for each subject between the two conditions and was then averaged across subjects. The resulting map was similar to the inter-subject CCM maps with the exception of the early visual cortex (V1), which demonstrated low concordance (see Figure 3).

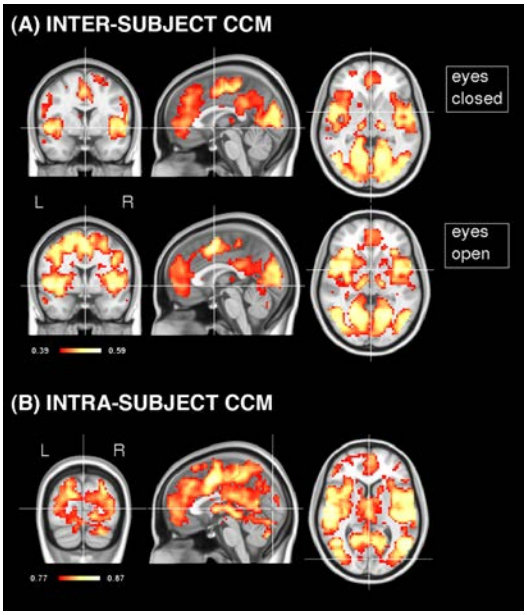

Figure 3: **CCM in healthy controls.**
(A) Inter-subject CCM for both experimental conditions (eyes open and eyes closed) depicts large proportions of the grey matter as highly concordant. Frontal areas and the default-mode network demonstrate lower concordance. (B) Intra-subject CCM computed between the two experimental conditions and then averaged across subjects. Note that early visual cortex (marked with crosshair) demonstrates lower concordance between the two experimental conditions. These results reflect divergent connectivity patterns between the two conditions captured using the CCM approach. Figure adapted from Study I (Lohmann, et al., 2012).

Experiment B:

CCM was computed for a single stroke patient who was examined repeatedly using rs-fMRI. When applied to all time points, CCM demonstrated larger concordance in grey matter, similar to healthy controls (experiment A). A local minimum of CCM was located in the left globus pallidus. When computing CCM for pairs of consecutive time points, the globus pallidus bilaterally was the area of greatest change when subtracting CCM(t2,t3) minus CCM(t3,t4). This suggests that the change in concordance in this area occurred at a later stage. Based on these findings, seed-based analysis was performed for both left and right globus pallidus for further exploration of the connectivity pattern driving this effect. CCM was hence used as a data-driven guidance for second-level analysis. Seed-based analysis was performed for the two latest time points. When subtracting the maps we found that the right globus pallidus generally demonstrated stronger connectivity to the rest of the brain than the left in the third time point, and this effect was reversed in the fourth time point after stroke (see Figure 4).

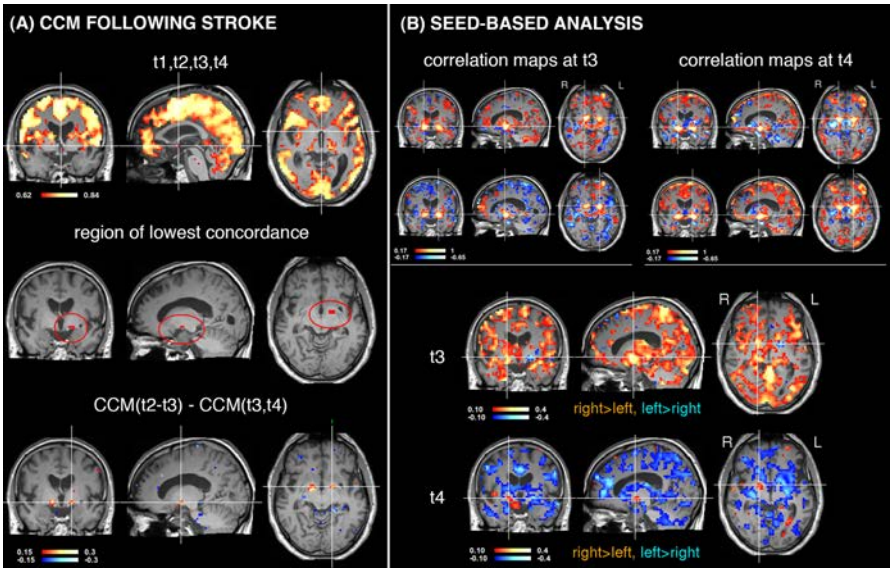

Figure 4: **CCM applied to longitudinal datasets following stroke.**
(A) Applying CCM to all four time points demonstrates large proportions of the grey matter as concordant. The region of lowest concordance marking the largest change in connectivity pattern over time is located in the left globus pallidus (middle panel). When computing CCM for pairs of consecutive time points, subtraction of these maps yields a large difference in concordance between the last two pairs of datasets located in bilateral globus pallidus. (B) Results based on CCM are used as a basis for second-level seed-based analysis. Functional connectivity was computed for both right and left globus pallidus (seed is marked with crosshair, see upper panels). Bottom row presents the difference in connectivity patterns for the third and the fourth time points for both seeds. During the third time point, connectivity is higher from the right globus pallidus to most regions, while during the fourth time points this pattern is reversed. Figure is adapted from Study I (Lohmann, et al., 2012).

5.1.4 Discussion

In Study I, we have presented the CCM algorithm and applied it to rs-fMRI data from healthy controls and a longitudinal dataset of a single stroke patient. In healthy controls, in line with our hypothesis, CCM differentiated the two experimental conditions (eyes open/eyes closed) such that CCM was lower in primary visual area (V1) in between the two states. This result replicates previous findings concerning divergent connectivity pattern within the visual cortex for rs-fMRI data based on eyes open and eyes closed (McAvoy, et al., 2008; Van Dijk, et al., 2010), and therefore supports the functional significance of CCM

and provides a proof of concept for future use of this novel algorithm. Additionally, in accordance with our expectations, inter-subject CCM in the healthy controls group detected most of grey matter areas as concordant while white matter voxels demonstrated low concordance. Areas of larger anatomical variability, such as frontal regions, demonstrated decreased concordance.

While we expected decreased concordance in frontal regions, it is important to emphasize that areas of greater inter-subject anatomical variability can lead to lower CCM values regardless of concordance of connectivity patterns. Although this problem is not specific to CCM and exists also in conventional General Linear Model (GLM) designs (K. J. Friston et al., 1994), which are widely used in the field of fMRI, a solution would be to apply CCM to longitudinal datasets from individual subjects. We pursued this approach on a clinical dataset from a single patient following stroke. The patient was scanned four consecutive times starting 1 day after the stroke. CCM was computed for all four scans and for pairs of consecutive scans. The left globus pallidus demonstrated minimal concordance values over time, pointing towards a large change in connectivity in this area. Based on the CCM results, seed-based analysis explored the change in connectivity structure leading to lower concordance values in the globus pallidus bilaterally. We found that the change in connectivity was attributed to a latter stage following the stroke (i.e. the third and forth time points) with an inter-hemispheric imbalance, as connectivity from the left globus palldius was stronger than the right in the third time point. This effect was reversed in the fourth time point. The globus pallidus is known to be part of the extrapyramidal motor system affected by the patient's thalamic stroke. Results from the CCM analysis suggest that the recovery process involved this node of the network, thus supporting the theoretical model of diaschisis. While these results require further validation in a larger cohort and the use of behavioral measures to strengthen the functional significance of our findings, we suggest that concordance can be used as a measure of longitudinal changes in connectivity to explore plasticity and recovery processes after stroke. On a more general note, CCM provides a powerful tool for the data-driven exploration of rs-fMRI data at the voxel level in order to guide second-level analyses.

5.2 Study II: Longitudinal effects of lesions on functional networks after stroke (Ovadia-Caro, et al., 2013)

5.2.1 Background

Study II investigates the distal effects of focal lesions on functional networks in longitudinal datasets following stroke. Changes in rs-fMRI functional connectivity over time were explored at the network level, taking into account patients with heterogeneous lesions affecting more than one functional network. The relationship between changes in functional connectivity and the behavioral trajectory was additionally tested.

Changes in rs-fMRI connectivity have been previously reported after stroke in single networks such as the sensory, motor, default-mode and attention (Carter, et al., 2010; Carter, Patel, et al., 2012; Golestani, et al., 2013; He, Snyder, et al., 2007; Park, et al., 2011; Tuladhar, et al., 2013; van Meer, et al., 2012; van Meer, van der Marel, Otte, et al., 2010; van Meer, van der Marel, Wang, et al., 2010; L. Wang, et al., 2010). These changes have been shown to correlate with the severity of behavioral deficits supporting the clinical significance of this approach. Exploring changes in functional connectivity in single networks was previously done by grouping patients based on similarity of lesion location or similarity of functional deficits in a single domain. However, stroke rarely affects only one functional domain or functional network. Additionally, no lesion is exactly the same in size and in location in two different patients. A unified model that can be applied to patients with different lesion locations and set of symptoms that may affect more than one network is needed. Based on the theoretical model of diaschisis (Andrews, 1991; Finger, et al., 2004; von Monakow, 1914, 1969), computational models (Alstott, et al., 2009; Honey & Sporns, 2008), and empirical results obtained in single networks (Carter, et al., 2010; Carter, Patel, et al., 2012; Golestani, et al., 2013; He, Snyder, et al., 2007; Park, et al., 2011; Tuladhar, et al., 2013; van Meer, et al., 2012; van Meer, van der Marel, Otte, et al., 2010; van Meer, van der Marel, Wang, et al., 2010; L. Wang, et al., 2010) we hypothesized that networks affected by the lesion (i.e. regions that are functionally connected to the lesion) would demonstrate a preferential

change in functional connectivity over time as compared to unaffected networks. In addition, the degree of change in functional connectivity should correlate with the behavioral trajectory in these patients.

5.2.2 Methods

Ischemic stroke patients (N=12, post-exclusion) received an rs-fMRI scan at day 1, day 7 and day 90 post-stroke. Following standard preprocessing, the lesions were defined such that a lesion mask was manually drawn based on the DWI or fluid attenuated inversion recovery (FLAIR) images of each patient. In order to determine which networks have been affected by the individual lesions, we computed an overlap between the lesion masks and a template set of eight networks previously published by Beckmann and colleagues (Beckmann, et al., 2005). This template of eight networks was computed using ICA applied to rs-fMRI data acquired in healthy controls. These eight networks include sensorimotor, visual, auditory and default-mode networks. Our choice of this template was based on the high functional relevance in addition to the high inter-subject consistency reported for these components (Beckmann, et al., 2005; Laird et al., 2011). As a result of computing an overlap between the eight networks template and the individual lesions, we defined for each subject which networks were affected by the lesion.

rs-fMRI data was analyzed using dual-regression analysis (Beckmann, et al., 2009; Filippini, et al., 2009). The a priori defined eight-network probabilistic maps served as regressors. As a result, each subject had 8 connectivity maps for each single scan. Based on the approach presented in study I (Lohmann, et al., 2012), we used the measure of concordance correlation coefficient to determine the degree of change in the spatial pattern of functional connectivity over time. Concordance was computed for each network separately over all time points, resulting in eight concordance values for each patient. Importantly, prior to concordance computation, we removed the lesion masks from the analysis. This was done because we were interested in the indirect connectivity effects induced by the lesion (i.e. diaschisis effect) and not the direct, local effects. In order to determine whether connectivity changed more in the networks affected by the

lesion, we computed delta-concordance for each subject such that average concordance values in unaffected networks were subtracted from average concordance values in affected networks ((μ unaffected) - (μ affected)). To test for significant differences between concordance in affected and unaffected networks we used a one-sample t-test (see Figure 5 for analysis steps).

In order to explore the relationship between connectivity changes (measured by concordance) and the behavioral trajectory, we used the National Institutes of Health Stroke Scale (NIHSS) scores from day 1 and day 90. Specifically, we computed a delta-NIHSS score defined as the ranked absolute difference between the NIHSS obtained at day 1, and the NIHSS obtained at day 90. Higher values represent a larger clinical change over time. Delta-NIHSS was correlated with delta-concordance using Spearman's correlation coefficient.

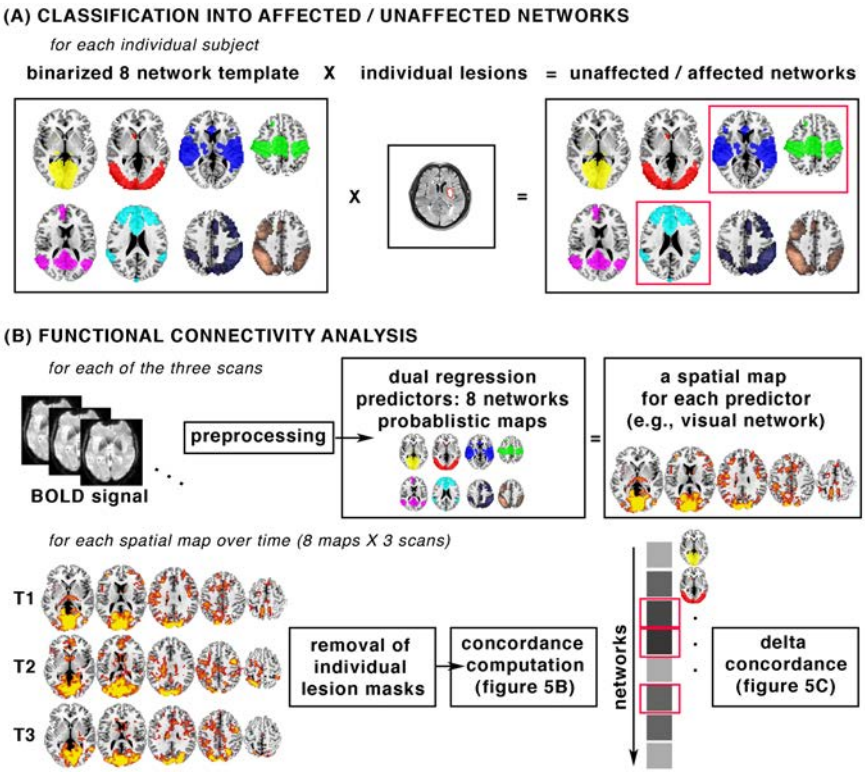

Figure 5: **Assessment of connectivity changes in multiple networks; a schematic illustration of analysis steps.**

(A) Classification of individual lesions into affected (red frame) and unaffected networks. The binarized eight- network template was multiplied by each individual lesion mask to classify lesions to affected/unaffected networks. (B) Functional connectivity analysis. Each scan was preprocessed and dual-regression was performed. After removal of individual lesion masks, spatial concordance correlation coefficient was computed for each map over three time points. Delta concordance ((μ unaffected)-(μ affected)) was computed and a one-sample t-test was performed.

5.2.3 Results

The impact of individual lesions on functional connectivity was computed using spatial concordance computed individually for each network, across all time points. Networks with high concordance values represent a small change in the spatial structure of connectivity maps, while networks with low concordance value represent a larger change. Based on our hypothesis, we expected that

delta-concordance would yield positive values. We found a significant difference ($p=0.018$, one-sided) indicating lower concordance in affected networks as compared to unaffected networks. Since the lesion area was excluded from this analysis, the effect found is reflecting a stronger indirect effect (i.e. diaschisis effect) of the lesion on functional connectivity in affected networks versus unaffected networks.

The clinical significance of our connectivity findings was tested based on the correlation between the behavioral trajectory as defined by delta-NIHSS and the change in functional connectivity over time as defined by delta-concordance. Based on previous studies (Carter, et al., 2010; He, Snyder, et al., 2007; Park, et al., 2011; L. Wang, et al., 2010), a positive correlation is expected such that larger effect on functional connectivity would correlate with stronger behavioral change. We found a significant ($p=0.05$, one-sided) positive correlation ($r=0.5$) between delta-concordance and delta-NIHSS indicating that stronger connectivity change corresponds to stronger effect on the behavioral trajectory (see Figure 6 for results summary).

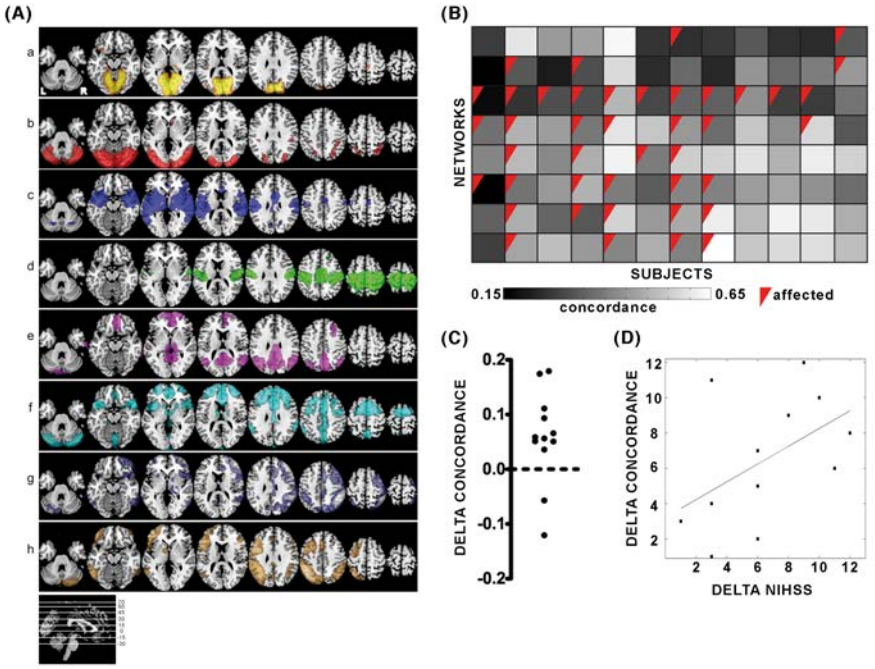

Figure 6: **Connectivity is preferentially impaired in affected networks.**
(A) The eight-network template. Axial slices of the eight independent components based on probabilistic independent component analysis in healthy controls (N=10) adopted from Beckmann, et al., 2005. This template was used as a basis for computation of overlap between lesions and networks as well as for dual-regression analysis. (B) Spatial concordance in affected and unaffected networks as computed over time for each patient (x-axis) and for each network (y-axis). High values reflect a small change in the spatial pattern. Red triangles depict affected networks. (C) Delta concordance was computed for each patient demonstrating a significant positive distribution as tested by one-sample t-test. (D) Relationship between delta concordance and clinical change. Positive correlation between changes in clinical scores over time as measured by delta NIHSS (x-axis) and changes in functional connectivity as measured by delta concordance (y-axis). Both axes depict the ranked values (as Spearman's correlation was applied to statistically test the relationship). Black line depicts the fitted regression line. Figure was adapted from Study II, (Ovadia-Caro, et al., 2013).

5.2.4 Discussion

Study II presents a novel approach to study rs-fMRI connectivity changes following stroke. This approach can be applied to heterogeneous lesions and can be used to simultaneously explore multiple networks. While most previous studies explored single networks, our approach can be used to map connectivity changes in a multi-faceted manner, potentially better reflecting the complex

changes exerted by a local stroke on the whole-brain, global level. Gaining a more detailed understanding of connectivity changes following stroke has a potential effect on refining treatment in the form of stimulation techniques, which have been shown to normalize connectivity patterns and improve behavioral status (Alonso-Alonso, et al., 2007; Di Pino, et al., 2014; Grefkes & Fink, 2011; Grefkes, et al., 2010; Sehm, et al., 2012).

We found that indirect effects of a local lesion on functional connectivity are more pronounced in affected networks, supporting the generalizability of connectivity changes reported previously in single networks (Carter, et al., 2010; Carter, Patel, et al., 2012; Golestani, et al., 2013; He, Snyder, et al., 2007; Park, et al., 2011; Tuladhar, et al., 2013; van Meer, et al., 2012; van Meer, van der Marel, Otte, et al., 2010; van Meer, van der Marel, Wang, et al., 2010; L. Wang, et al., 2010). Starting with data acquired 1 day after stroke, a significant decrease in concordance was found for networks containing lesions compared with unaffected networks. Our results are in line with the theoretical model of diaschisis suggesting that stroke induces indirect changes in structurally intact areas connected to it (Andrews, 1991; Finger, et al., 2004; von Monakow, 1914, 1969). Importantly, the measure of concordance was correlated with post-stroke behavioral trajectory thus supporting the potential clinical significance of our findings. Our results are hence supportive of both our initial hypotheses.

5.3 Study III: The value of resting-state functional magnetic resonance imaging in stroke (Ovadia-Caro, et al., 2014)

rs-fMRI has been successfully applied to map the functional organization of the brain in healthy subjects (Sporns, et al., 2005) as well as to study various clinical populations (M. D. Fox & Greicius, 2010). In recent years, rs-fMRI has also been used to study stroke pathology and plasticity (Carter, Shulman, et al., 2012). Being a relatively new approach, study III aimed at reviewing all 17 studies published thus far in the field, discussing their main results and concluding on the clinical utility of this approach. All studies are summarized in supplementary Table 1 of the paper (see full-text PDF files).

5.3.1 Background

The purpose of using imaging techniques following stroke varies depending on the time that has passed since stroke onset. In the acute phase, imaging is aimed at gathering information concerning the vascular patency, areas of hypoperfusion and metabolic and structural damage in order to guide therapeutic decisions such as reperfusion therapies. In the sub-acute and chronic phase, reorganization and plasticity take place in order to regain impaired functions. In this phase, reorganization of distributed networks mediated by connectivity is taking place (Andrews, 1991). Imaging is hence aimed at exploring the status of cerebral networks and the interaction between regions within affected networks. rs-fMRI is an ideal imaging approach to be used for this purpose as it allows for multiple cerebral network assessment in a single scan.

5.3.2 Using rs-fMRI in the acute phase as a measure of local perfusion

The BOLD signal contains information concerning local blood flow and oxygen consumption in addition to information concerning neuronal activity (Villringer & Dirnagl, 1995; Vincent, et al., 2007). While in common rs-fMRI connectivity analyses, the aim of preprocessing is to minimize the effect of vascular signals, in the acute phase after stroke this component of the BOLD signal could prove beneficial to identify potentially salvable tissue on appropriate therapy. Recent

applications of rs-fMRI in acute stroke have demonstrated that it may be used to replace perfusion measurements that require contrast agent (Amemiya, et al., 2013; Lv, et al., 2013). The main limitations of contrast agent usage are the inability to repeat the measurement in case of motion artifacts, and severe side effects such as nephrogenic systemic fibrosis. Using time-shift analysis, two studies have recently reported high spatial similarity between areas of hypoperfusion defined on perfusion imaging and areas of delayed BOLD signal defined using rs-fMRI (Amemiya, et al., 2013; Lv, et al., 2013). Although these results are preliminary due to the small number of subjects, they suggest a promising diagnostic usage for rs-fMRI in the acute phase.

5.3.3 Using rs-fMRI in the sub-acute and chronic phase to study reorganization

Following the acute phase, alterations in rs-fMRI connectivity can be used to explore reorganization of functional networks. rs-fMRI signal provides information concerning the interaction between distant regions, an analysis known as functional connectivity (K. J. Friston, 2011). The main finding reported after stroke in both animal (van Meer, et al., 2012; van Meer, van der Marel, Otte, et al., 2010; van Meer, van der Marel, Wang, et al., 2010) and human studies (Carter, et al., 2010; Carter, Patel, et al., 2012; Golestani, et al., 2013; He, Snyder, et al., 2007; Nomura, et al., 2010; Park, et al., 2011; Tuladhar, et al., 2013; L. Wang, et al., 2010) is that functional connectivity is reduced in areas structurally intact yet connected to the lesion area. Decreased connectivity has been reported for single networks among them motor, attention and default-mode. The reduction in connectivity was associated with impaired function in the corresponding behavioral domain further supporting the clinical significance of these findings (Carter, et al., 2010; Golestani, et al., 2013; He, Snyder, et al., 2007; Park, et al., 2011; van Meer, et al., 2012; van Meer, van der Marel, Wang, et al., 2010; L. Wang, et al., 2010). Stroke is hence causing a network-level disruption other than the localized effects in the vicinity of the infarct. This view of stroke as a network disorder along with the recent methodological and computational advancements in the field have caused a shift in the methodologies applied to

study reorganization after stroke. The aim is to develop experimental approaches that can assess multiple network damage in patients with heterogeneous lesions while maximizing the usage of raw data (i.e. whole-brain analyses). Our work (presented here as study I and II) suggests the usage of concordance to quantify changes in functional connectivity patterns over time on the whole brain level (Lohmann, et al., 2012). We found that the diaschisis-effects reported for single networks are evident also when assessing multiple networks, and have shown that the degree of change in functional connectivity over time correlated with the behavioral trajectory in individual patients further supporting the potential prognostic value of rs-fMRI after stroke (Ovadia-Caro, et al., 2013).

Additional developments in the field have suggested the application of methods from the mathematical field of graph theory to rs-fMRI data. A graph is defined as a set of nodes (corresponding to brain regions or voxels) and edges (corresponding to connections between the nodes). In the case of rs-fMRI, connections are usually reflected by correlation strength between signals from different nodes. Graph theory measures can inform us on the integrative ability of different regions within the network and can provide information on the topological organization of single as well as multiple networks. Such measures include centrality, path length, clustering coefficient and modularity. These measures can quantify the effectiveness and structural nature of the network under investigation (Bullmore & Sporns, 2009). Graph theory has been applied to explore network structure within the motor networks in both humans (L. Wang, et al., 2010) and animals (van Meer, et al., 2012) after stroke. Additionally, the role of the lesion topology within the network was explored using this approach, and lesions falling within areas connecting different networks (i.e. connector hubs), have been demonstrated to cause a greater impairment to the network integrity as measured by modularity (Gratton, et al., 2012). Although graph theory is a recent and increasingly popular application to rs-fMRI data, today's computing power requires a reduction in the number of areas to be analyzed through parcellation methods (Craddock, et al., 2012; Tzourio-Mazoyer, et al., 2002). With further developments in computational capabilities, graph theory approach will be applied using larger number of regions, thereby creating

more realistic graphs. In addition, the link between graph-based measures and recovery after stroke should be further explored to conclude on the clinical utility of this approach.

5.3.4 Limitations and considerations

As the application of rs-fMRI to study stroke is relatively recent, several limitations need to be addressed by future studies. In particular, while some rs-fMRI connectivity analyses are more susceptible to motion artifacts than others, the need for good post hoc removal of motion artifacts or real-time motion correction is still evident (K. Murphy et al., 2013; Power et al., 2012). Furthermore, structural damage of the white matter has been shown to impair connectivity (J. S. Damoiseaux & Greicius, 2009; A Schaefer, Quinque, et al., 2014) and to account for some of the behavioral deficits observed after stroke (Johansen-Berg et al., 2010). Since white matter lesions are very common in stroke patients, in part due to their age, this variable should be reported and accounted for either by excluding patients with severe white matter lesions or by means of regression.

The status of vascular pathology and local changes in perfusion and metabolism may also affect the connectivity analysis. While local changes in perfusion are most prominent in the first hours after stroke and most studies conducted in the field use data acquired later on, the vascular pathology (e.g., stenosis) is in some cases a pre-existing state and poses some difficulties in interpreting the source of observed connectivity changes. The BOLD signal is an indirect measure of neuronal activity measuring changes in the concentration of deoxyhemoglobin (Logothetis, 2003; Villringer & Dirnagl, 1995). The interaction between the neuronal part and vascular part is based on complex mechanisms of neurovascular coupling (Villringer & Dirnagl, 1995). However, it is unclear whether similar neurovascular coupling can be assumed between healthy controls and stroke patients with local ischemia and in most cases pre-existing vascular disease such as stenosis (D'Esposito et al., 2003). While there are reports in the literature of changes in the BOLD signal resulting from mere vascular change (Krainik et al., 2005; Mazzetto-Betti et al., 2010; Pineiro et al.,

2002), the effect on rs-fMRI connectivity analysis has not been explored yet. Individual level analysis and correlating connectivity changes to behavioral improvement can minimize this problem and support a neuronal origin for the observed changes.

5.3.5 Conclusions

In this review paper, we have described and discussed the potential usage of rs-fMRI to stroke patients in the acute and chronic phase. In the acute stage, rs-fMRI has been found to be of potential diagnostic value by describing the perfusion deficit. In the sub-acute and chronic phase, rs-fMRI connectivity analysis is being used to explore reorganization and plasticity. Using this approach, local stroke has been shown to cause a network wide disruption in the involved network, and to result in whole-brain topological changes. Connectivity changes have been shown to correlate with various behavioral measures, further supporting the prognostic value of rs-fMRI and the usage of connectivity as a basis for guided therapy in the form of stimulation techniques (Alonso-Alonso, et al., 2007; Di Pino, et al., 2014; Grefkes & Fink, 2011; Grefkes, et al., 2010; Sehm, et al., 2012).

6 General discussion

6.1 summary of results

The main goal of the current dissertation was to explore plastic changes following stroke as measured by rs-fMRI data. We propose a methodological and conceptual framework to study stroke pathology as a *network disruption* rather than a mere localized phenomenon.

Study I presented a novel, data-driven mathematical tool to analyze rs-fMRI data named connectivity concordance mapping (CCM) (Lohmann, et al., 2012). Specifically, we quantify the reproducibility of functional connectivity patterns at the whole-brain voxel level using a measure of connectivity concordance. This study describes the algorithm in detail and presents results from applying the method to healthy controls data as well as a longitudinal dataset from a single stroke patient.

Our results from the healthy controls sample are in line with previous findings demonstrating divergent connectivity patterns within the visual cortex during eyes open and eyes closed conditions (McAvoy, et al., 2008; Van Dijk, et al., 2010), supporting the functional significance of the algorithm and providing a proof of concept for its use. When applying CCM to the longitudinal dataset of a patient following ischemic stroke in the left thalamus, the bilateral globus pallidus demonstrated decreased concordance over time. Given the well-established structural links between the thalamus and globus pallidus these results are supportive of the theoretical model of diaschisis. However, considering the lack of behavioral measures supporting our findings, this result should be used as a catalyst for more in-depth exploration in larger samples (as provided in study II).

The application of CCM to rs-fMRI data is an exciting new tool with applicability to rs-fMRI data from both healthy controls and patients' data. We suggest concordance as a quantitative measure of plasticity as reflected in changes of the spatial pattern of functional connectivity maps. Concordance has been previously used to measure reproducibility in fMRI experiments (B. B. Biswal, et al., 2010; Hasson, et al., 2010; Lange et al., 1999; Shehzad, et al., 2009). However, none of

these applications was performed at the whole-brain voxel level. CCM can be further used as a data-driven approach to detect areas of interests for second-level analyses.

Study II explored the distal effects of lesions on functional network connectivity patterns. Starting at day 1 after stroke, longitudinal rs-fMRI data from a heterogeneous group of patients with varying lesions were collected. While most studies conducted thus far in the field explore single networks with a relatively small number of regions, we have presented an analytic approach taking into account multiple functional networks. This approach can be applied to individual-level analysis, and hence can be used to study patients with heterogeneous lesions affecting more than one network. We have explored the change over time in the spatial connectivity pattern of these networks using the measure of concordance (initially suggested in study I).

We found that networks affected by the lesion presented a preferential change in the spatial connectivity pattern over time, supporting the theoretical model of diaschisis. In addition, we have determined a link between the degree of change and the degree of behavioral recovery, further supporting the functional significance of our empirical findings and their potential prognostic value (Ovadia-Caro, et al., 2013). Our results are in line with previous findings obtained using single network analysis in the motor (Carter, et al., 2010; Carter, Patel, et al., 2012; Golestani, et al., 2013; Park, et al., 2011), attention (Gillebert et al., 2011; He, Snyder, et al., 2007), and default-mode networks (Lassalle-Lagadec et al., 2012; Tuladhar, et al., 2013). Similar results have been reported for experimental stroke in animal studies within the sensorimotor network (van Meer, et al., 2012; van Meer, van der Marel, Otte, et al., 2010; van Meer, van der Marel, Wang, et al., 2010) and in computational models simulating the effects of lesions on network topology (Alstott, et al., 2009; Cabral et al., 2012; de Haan et al., 2012; Honey & Sporns, 2008). All these studies taken together suggest that localized lesions cause widespread effect to interconnected areas, limited by the boundaries of the affected network.

Preferential change in functionally affected networks has been previously reported in patients with heterogeneous lesions affecting one of two networks. This work was conducted on patients in the chronic phase (at least 5 months

post-ictus) using a single rs-fMRI scan with the main goal of testing the dissociation of two cognitive control networks (Nomura, et al., 2010). Our work was inspired by this study and extends these findings to study recovery processes in multiple functional networks. We suggest the multi-network assessment of lesions' distal effects as a unified model to study all stroke patients.

6.2 Practical and theoretical implications

Our work has both practical and theoretical implications. On the clinical aspect, a unified model that takes multiple network damage into account has the potential of better representing complex clinical symptoms after stroke involving more than one functional domain. Given the correlation between changes in functional connectivity over time and the behavioral trajectory, our individual-level analysis may provide a framework for individualized prognosis in patients with heterogeneous lesions. Importantly, therapeutic attempts using non-invasive stimulation techniques are currently being performed in single functional domains, mainly in the motor network (Grefkes & Fink, 2011, 2012; Grefkes & Ward, 2014; Hummel & Cohen, 2006). TMS and transcranial direct current stimulation (tDCS) have been shown to successfully modulate connectivity in cerebral networks (Grefkes & Fink, 2011; Sehm, et al., 2012). Non-invasive stimulation techniques are used to normalize connectivity changes induced by the stroke to improve function (Alonso-Alonso, et al., 2007; Di Pino, et al., 2014; Grefkes & Fink, 2011; Grefkes, et al., 2010; Sehm, et al., 2012). However, results are mixed in terms of behavioral improvement (Ameli, et al., 2009; Grefkes & Ward, 2014). A multi-network assessment of changes in functional connectivity can be used to tailor stimulation protocols at the individual level thereby potentially improving the efficiency of this exciting therapeutic approach.

On the theoretical level, stroke provides a unique model of a localized structural damage to a system that is functional prior to injury (Carter, Shulman, et al., 2012). Unlike other progressive neurological diseases, any alterations in the system that follow a sudden stroke reflect reorganization within the network in the aim of regaining homeostasis. The lesion can be viewed as a knockout of a

single node in the network causing a widespread dysfunction. Stroke can hence inform us on various basic aspects of intrinsic network organization that apply to healthy brains as well.

Our results support the independence between different functional networks since lesions had a preferential distal effect in affected networks as compared to unaffected networks. The decreased concordance over time in affected networks provides empirical support for one of the fundamental principles underlying brain function, specifically as it pertains to information flow. In order to produce effective function, the brain has to be able to process information within single modules as well combine information coming from different modules. Successful function is achieved through a constant, dynamic balance between what is commonly referred to as *segregation* and *integration* of information (Deco et al., 2015; Sporns, 2013). This process is essential for functioning of distributed networks underlying cognitive functions (P. T. Fox & Friston, 2012; Tononi et al., 1994).

Results from network analyses using tools from the mathematical field of graph theory suggest network communities account for segregation, while inter-network hubs account for integration (Deco, et al., 2015; Sporns, 2013). Network communities are defined by higher connectivity within specific networks/modules and a certain degree of independence between different networks (Bullmore & Sporns, 2009). Our results are hence supportive of an underlying network structure promoting segregation of information.

In order to produce complex cognitive functions involving more than one modality, information has to be integrated as well. This is achieved through inter-network hubs; regions of disproportionate connectivity binding information between two (or more) communities (Deco, et al., 2015; Sporns, 2013).

This dual description of the network is commonly referred to as 'small-world' topology (Watts & Strogatz, 1998); on the one hand nodes are highly clustered locally, yet on the other hand, the average path length between two nodes of the network is relatively short due to hub regions (Bullmore & Sporns, 2009). 'Small-world' topology was originally described for social networks (Milgram, 1967), yet multiple evidences for 'small-world' properties have been reported

both at the whole-brain scale using neuroimaging data, and at cellular level using data acquired in animal models (Bassett & Bullmore, 2006; Bullmore & Sporns, 2009; Reijneveld et al., 2007; Sporns et al., 2004; Stam & Reijneveld, 2007) supporting an infrastructure for cost effective segregation and integration of information in the brain (Achard & Bullmore, 2007; Bullmore & Sporns, 2012).

The idea that different functional networks are segregated can be additionally used as basis to test different neurocognitive models established in healthy subjects. Nomura and colleagues explored the influence of lesions distributed in two different networks, the fronto-parietal and cingulo-opercular, on functional connectivity in order to demonstrate the dissociation of these two cognitive control networks (Nomura, et al., 2010). A similar approach was employed by He and colleagues within the attention domain (Corbetta, et al., 2005; He, Shulman, et al., 2007).

To summarize, in addition to the clinical utility, empirical evidence for the impact of lesions on functional networks provide further knowledge of the network structure as well as topological properties that may shed light on basic principles such as segregation and integration of information in the healthy brain.

6.3 Considerations and suggestions for further research

Our suggested multi-network approach to study plasticity in stroke patients is a novel approach providing a solution for the challenge of heterogeneity in the form of a unified model that can be used in all stroke patients. However, some limitations of our methodology should be taken into account.

The measure of concordance correlation coefficient to quantify changes in functional connectivity pattern ignores the directionality and source of change. The underlying nature of connectivity changes should hence be further explored with additional second-level analyses (such as was done in study I using seed-based analysis), in order to detect specific areas that contribute to the decreased concordance. The idea that some networks are more innately concordant than others (Lohmann, et al., 2012; Shehzad, et al., 2009) should also be taken into consideration. However, concordance was computed in our analysis for each subject individually and was normalized by concordance values in unaffected

networks. The heterogeneity of lesions in our sample further minimized the potential confound of within-subject comparison.

The correlation between changes in functional connectivity following stroke and behavioral impairments have been previously reported for single networks, and their corresponding functional domains (Carter, et al., 2010; He, Snyder, et al., 2007; Park, et al., 2011; L. Wang, et al., 2010). In study II we found a relationship between the degree of change in functional connectivity over time and the degree of behavioral change over time as measured by the National Institutes of Health Stroke Scale (NIHSS). While these results strengthen the clinical significance of our findings, NIHSS is a fairly general measure of dysfunction after stroke that mainly assesses motor function. Future studies should explore the relationship between specific clinical outcome involving multiple networks and their corresponding behavioral domains and concordance. Assembling a battery of behavioral tests that match each assessed network would further increase the clinical utility and prognostic value of our approach.

The impact of lesions within the white matter on functional connectivity has been previously demonstrated (J. S. Damoiseaux & Greicius, 2009; Johansen-Berg, et al., 2010; A Schaefer, et al., 2014). In our sample, we excluded subjects with moderate to severe white matter lesions (measured as Wahlund score ≥ 6) in order to avoid this confound. However, a model that is aimed at optimal characterization of symptoms should take the contribution of this factor into account. Such a model would be applicable to a larger population of stroke patients thereby increasing its clinical utility.

Understanding the distributed effects caused by a stroke can be further explored based on the concepts of segregation and integration of information and the influence of the lesion on the network topology. Computational modeling suggests that in addition to the widespread effects caused by the lesion, the topological role of the lesion itself within the network can partially account for the degree to which the network is impaired. That is, simulated lesions located in areas of high connectivity (i.e. hub regions) had a more detrimental effect on alterations in functional connectivity (Alstott, et al., 2009; de Haan, et al., 2012; Honey & Sporns, 2008). Recently, the role of lesion topology in alterations of functional connectivity after stroke has been explored empirically. Gratton and

colleagues have shown that lesions located in hub regions had a more detrimental effect on network integrity measured by modularity. Additionally, they have demonstrated that the association between hub damage and modularity could not be explained by the lesion size alone. This study demonstrates the importance of hub regions to the integrity of the network (Gratton, et al., 2012). Although the link between hub damage and behavior is yet to be explored, the topology of the lesion site provides an important insight on the widespread network damage that could prove to be clinically relevant in the future.

To date, studying the effects of lesions has been done using stationary measures of functional connectivity. That is, the rs-fMRI data from the complete scan is used in order to compute correlation between pairs of regions. Such static analysis captures in essence only a snapshot of the effects of the stroke on the network. The delicate balance between segregation and integration of information is a dynamic process, hence suggesting that changes in functional connectivity, and in network topology during the scan are of functional relevance (Hutchison, et al., 2013; Sporns, 2013). Recent studies have documented significant fluctuations of connectivity across different time-scales ranging from milliseconds to seconds in both task-based (Ekman et al., 2012; Fornito et al., 2012; Kinnison et al., 2012) and rs-fMRI data from both animals and humans (Chang & Glover, 2010; Handwerker et al., 2012; Hutchison, Womelsdorf, Gati, et al., 2013; Keilholz et al., 2013) pointing at the behavioral significance of this approach. It has been additionally suggested that changes in functional connectivity observed in clinical populations such as Alzheimer's can be accounted for by nonstationary temporal connectivity changes (Jones et al., 2012). Windowing of rs-fMRI data to explore network dynamics is a newly emerging frontier in the study of brain connectivity (Hutchison, et al., 2013; Sporns, 2013). Although network dynamics require longer scans which could be challenging in patients after stroke, such analysis may shed light on the underlying process leading to widespread reduction in functional connectivity and can inform us on the dynamic nature of both segregation and integration in these unique conditions.

More specifically, following stroke, hub regions play a central role in network structure and integrity as explored by both human studies (Gratton, et al., 2012) and computational models (Alstott, et al., 2009; de Haan, et al., 2012; Honey & Sporns, 2008) using stationary connectivity measures. Since hub regions connect different networks, it is assumed that over time, hubs will vary their degree of connectivity to one network over the other based on changing cognitive needs. These alterations are likely to reflect the ongoing integration process of information flow. Recently it has been shown that using rs-fMRI data and a connectivity clustering approach, hubs can be part of multiple networks and their degree of integration into one network over the other changes over time. Moreover, the same study showed that the degree of variation in hub integration to one network over the other was decreased with age and was related to stimulus independent thoughts during the scan (Schaefer et al., 2014). These results point at a possible cognitive role for dynamic changes in the degree of hubs integration. Similarly, additional studies have shown that dynamic changes in modularity of the network play a role in learning and mood changes over multiple time scales (Bassett et al., 2011; Betzel et al., 2016). Owing to the role hubs play in integration of information in the healthy brain, it can be assumed that these areas are involved in the reorganization process following stroke. However, the link between hubs dynamic integration, functional connectivity changes and behavioral recovery after stroke has not been explored. Such analysis could provide an interesting future direction to the study of reorganization and changes in integration abilities within the networks following stroke.

6.4 Conclusions

This dissertation sought to explore post-stroke network reorganization and the recovery of functional networks as measured by rs-fMRI. In three studies summarized here, this goal was approached from slightly different angles. Presenting novel methodological approaches to rs-fMRI stroke data as well as empirical findings to support the current view of stroke as a network disruption rather than a localized structural phenomenon, and ending with a general

overview of the field and the possible utility, clinical significance and limitations of this experimental approach. In accordance with previous findings obtained in single networks and the theoretical model of diaschisis, our results support the notion that stroke should be understood, explored and ideally treated as a network disruption, and that areas connected to the lesion are of paramount functional relevance to the recovery process following stroke.

References

Achard, S., & Bullmore, E. (2007). Efficiency and cost of economical brain functional networks. *PLoS computational biology, 3*(2), e17.

Alonso-Alonso, M., Fregni, F., & Pascual-Leone, A. (2007). Brain stimulation in poststroke rehabilitation. *Cerebrovascular diseases, 24 Suppl 1*, 157-66.

Alstott, J., Breakspear, M., Hagmann, P., Cammoun, L., & Sporns, O. (2009). Modeling the impact of lesions in the human brain. *PLoS computational biology, 5*(6), e1000408.

Ameli, M., Grefkes, C., Kemper, F., Riegg, F. P., Rehme, A. K., Karbe, H., et al. (2009). Differential effects of high-frequency repetitive transcranial magnetic stimulation over ipsilesional primary motor cortex in cortical and subcortical middle cerebral artery stroke. *Annals of neurology, 66*(3), 298-309.

Amemiya, S., Kunimatsu, A., Saito, N., & Ohtomo, K. (2013). Cerebral Hemodynamic Impairment: Assessment with Resting-State Functional MR Imaging. *Radiology*.

Anand, A., Li, Y., Wang, Y., Wu, J., Gao, S., Bukhari, L., et al. (2005). Activity and connectivity of brain mood regulating circuit in depression: a functional magnetic resonance study. *Biol Psychiatry, 57*(10), 1079-88.

Andrews, R. J. (1991). Transhemispheric diaschisis. A review and comment. *Stroke; a journal of cerebral circulation, 22*(7), 943-9.

Arieli, A., Shoham, D., Hildesheim, R., & Grinvald, A. (1995). Coherent spatiotemporal patterns of ongoing activity revealed by real-time optical imaging coupled with single-unit recording in the cat visual cortex. *Journal of neurophysiology, 73*(5), 2072-93.

Arieli, A., Sterkin, A., Grinvald, A., & Aertsen, A. (1996). Dynamics of ongoing activity: explanation of the large variability in evoked cortical responses. *Science, 273*, 1868-71.

Bar, M. (2007). The proactive brain: using analogies and associations to generate predictions. *Trends in cognitive sciences, 11*(7), 280-9.

Bar, M. (2009). The proactive brain: memory for predictions. ***Philosophical transactions of the Royal Society of London Series B, Biological sciences, 364*****(1521), 1235-43.**

Bassett, D. S., & Bullmore, E. (2006). Small-world brain networks. ***Neuroscientist, 12*****(6), 512-23.**

Bassett, D. S., Wymbs, N. F., Porter, M. A., Mucha, P. J., Carlson, J. M., & Grafton, S. T. (2011). Dynamic reconfiguration of human brain networks during learning. ***Proceedings of the National Academy of Sciences of the United States of America, 108*****(18), 7641-6.**

Becker, R., Reinacher, M., Freyer, F., Villringer, A., & Ritter, P. (2011). How ongoing neuronal oscillations account for evoked fMRI variability. ***The Journal of neuroscience : the official journal of the Society for Neuroscience, 31*****(30), 11016-27.**

Beckmann, C. F., DeLuca, M., Devlin, J. T., & Smith, S. M. (2005). Investigations into resting-state connectivity using independent component analysis. ***Philosophical transactions of the Royal Society of London Series B, Biological sciences, 360*****(1457), 1001-13.**

Beckmann, C. F., Mackay, C. E., Filippini, N., & smith, S. (2009). Group Comparison of resting-state FMRI data using multi-subject ICA and dual regression. ***Fifteenth Annual Meeting of the Organization for Human Brain Mapping*****.**

Berger, H. (1929). Uber des Elektrenkephalogramm des Menschen. ***Arch Psychiatr Nervenkrankheiten, 87*****, 527-80.**

Berger, H. (1969). On the electroencephalogram of man. ***Electroencephalogr Clin Neurophysiol*****, Suppl 28:37+.**

Betzel, R. F., Satterthwaite, T. D., Gold, J. I., & Bassett, D. S. (2016). A positive mood, a flexible brain. ***arXiv:160107881v1*****.**

Birn, R. M. (2012). The role of physiological noise in resting-state functional connectivity. ***NeuroImage, 62*****(2), 864-70.**

Birn, R. M., Diamond, J. B., Smith, M. A., & Bandettini, P. A. (2006). Separating respiratory-variation-related fluctuations from neuronal-activity-related fluctuations in fMRI. ***Neuroimage, 31*****(4), 1536-48.**

Biswal, B., Yetkin, F. Z., Haughton, V. M., & Hyde, J. S. (1995). Functional connectivity in the motor cortex of resting human brain using echoplanar MRI. ***Magnetic Resonance Medicine, 34*****, 537-41.**

Biswal, B. B., Mennes, M., Zuo, X. N., Gohel, S., Kelly, C., Smith, S. M., et al. (2010). Toward discovery science of human brain function. ***Proceedings of the National Academy of Sciences of the United States of America, 107*****(10), 4734-9.**

Bluhm, R. L., Miller, J., Lanius, R. A., Osuch, E. A., Boksman, K., Neufeld, R. W., et al. (2007). Spontaneous low-frequency fluctuations in the BOLD signal in schizophrenic patients: anomalies in the default network. ***Schizophr Bull, 33*****(4), 1004-12.**

Boly, M., Massimini, M., Garrido, M. I., Gosseries, O., Noirhomme, Q., Laureys, S., et al. (2012). Brain connectivity in disorders of consciousness. ***Brain connectivity, 2*****(1), 1-10.**

Broca, P. (1861b). Perte de la parole: ramollissement chronique et destruction partielle du lobe anterieur gauche du cerveau. ***Bulletins de la Societe d'anthropologie, 1re serie, 2*****, 235-8.**

Brookes, M. J., Woolrich, M., Luckhoo, H., Price, D., Hale, J. R., Stephenson, M. C., et al. (2011). Investigating the electrophysiological basis of resting state networks using magnetoencephalography. ***Proceedings of the National Academy of Sciences of the United States of America, 108*****(40), 16783-8.**

Buckner, R. L. (2010). Human functional connectivity: new tools, unresolved questions. ***Proceedings of the National Academy of Sciences of the United States of America, 107*****(24), 10769-70.**

Buckner, R. L., Sepulcre, J., Talukdar, T., Krienen, F. M., Liu, H., Hedden, T., et al. (2009). Cortical hubs revealed by intrinsic functional connectivity: mapping, assessment of stability, and relation to Alzheimer's disease. ***J Neurosci, 29*****(6), 1860-73.**

Bullmore, E., & Sporns, O. (2009). Complex brain networks: graph theoretical analysis of structural and functional systems. *Nature reviews Neuroscience, 10*(3), 186-98.

Bullmore, E., & Sporns, O. (2012). The economy of brain network organization. *Nature reviews Neuroscience, 13*(5), 336-49.

Burke, R. E. (2007). Sir Charles Sherrington's the integrative action of the nervous system: a centenary appreciation. *Brain : a journal of neurology, 130*(Pt 4), 887-94.

Cabral, J., Hugues, E., Kringelbach, M. L., & Deco, G. (2012). Modeling the outcome of structural disconnection on resting-state functional connectivity. *NeuroImage, 62*(3), 1342-53.

Caleo, M. (2015). Rehabilitation and plasticity following stroke: Insights from rodent models. *Neuroscience, 311*, 180-94.

Carmichael, S. T. (2003). Plasticity of cortical projections after stroke. *The Neuroscientist : a review journal bringing neurobiology, neurology and psychiatry, 9*(1), 64-75.

Carmichael, S. T. (2006). Cellular and molecular mechanisms of neural repair after stroke: making waves. *Annals of neurology, 59*(5), 735-42.

Carmichael, S. T. (2008). Themes and strategies for studying the biology of stroke recovery in the poststroke epoch. *Stroke; a journal of cerebral circulation, 39*(4), 1380-8.

Carmichael, S. T., & Chesselet, M. F. (2002). Synchronous neuronal activity is a signal for axonal sprouting after cortical lesions in the adult. *The Journal of neuroscience : the official journal of the Society for Neuroscience, 22*(14), 6062-70.

Carmichael, S. T., Kathirvelu, B., Schweppe, C. A., & Nie, E. H. (2016). Molecular, cellular and functional events in axonal sprouting after stroke. *Experimental neurology*.

Carmichael, S. T., Saper, C., & Schlaug, G. (2016). Emergent properties of neural repair: Elemental biology to therapeutic concepts. *Annals of neurology*.

Carmichael, S. T., Wei, L., Rovainen, C. M., & Woolsey, T. A. (2001). New patterns of intracortical projections after focal cortical stroke. *Neurobiology of disease, 8*(5), 910-22.

Carrera, E., & Tononi, G. (2014). Diaschisis: past, present, future. *Brain : a journal of neurology, 137*(Pt 9), 2408-22.

Carter, A. R., Astafiev, S. V., Lang, C. E., Connor, L. T., Rengachary, J., Strube, M. J., et al. (2010). Resting interhemispheric functional magnetic resonance imaging connectivity predicts performance after stroke. *Annals of neurology, 67*(3), 365-75.

Carter, A. R., Patel, K. R., Astafiev, S. V., Snyder, A. Z., Rengachary, J., Strube, M. J., et al. (2012). Upstream dysfunction of somatomotor functional connectivity after corticospinal damage in stroke. *Neurorehabilitation and neural repair, 26*(1), 7-19.

Carter, A. R., Shulman, G. L., & Corbetta, M. (2012). Why use a connectivity-based approach to study stroke and recovery of function? *NeuroImage, 62*(4), 2271-80.

Chang, C., & Glover, G. H. (2010). Time-frequency dynamics of resting-state brain connectivity measured with fMRI. *NeuroImage, 50*(1), 81-98.

Chen, R., Cohen, L. G., & Hallett, M. (2002). Nervous system reorganization following injury. *Neuroscience, 111*(4), 761-73.

Cherkassky, V. L., Kana, R. K., Keller, T. A., & Just, M. A. (2006). Functional connectivity in a baseline resting-state network in autism. *Neuroreport, 17*(16), 1687-90.

Corbetta, M. (2012). Functional connectivity and neurological recovery. *Developmental Psychobiology, 54*(3), 239-53.

Corbetta, M., Kincade, M. J., Lewis, C., Snyder, A. Z., & Sapir, A. (2005). Neural basis and recovery of spatial attention deficits in spatial neglect. *Nature neuroscience, 8*(11), 1603-10.

Corbetta, M., & Shulman, G. L. (2011). Spatial neglect and attention networks. *Annual review of neuroscience, 34*, 569-99.

Craddock, R. C., James, G. A., Holtzheimer, P. E., 3rd, Hu, X. P., & Mayberg, H. S. (2012). A whole brain fMRI atlas generated via spatially constrained spectral clustering. ***Human brain mapping, 33*****(8), 1914-28.**

Cramer, S. C., Sur, M., Dobkin, B. H., O'Brien, C., Sanger, T. D., Trojanowski, J. Q., et al. (2011). Harnessing neuroplasticity for clinical applications. ***Brain : a journal of neurology, 134*****(Pt 6), 1591-609.**

D'Esposito, M., Deouell, L. Y., & Gazzaley, A. (2003). Alterations in the BOLD fMRI signal with ageing and disease: a challenge for neuroimaging. ***Nature reviews Neuroscience, 4*****(11), 863-72.**

Damoiseaux, J. S., & Greicius, M. D. (2009). Greater than the sum of its parts: a review of studies combining structural connectivity and resting-state functional connectivity. ***Brain Structure & Function, 213*****(6), 525-33.**

Damoiseaux, J. S., Rombouts, S. A. R. B., Barkhof, F., Scheltens, P., Stam, C. J., Smith, S. M., et al. (2006). Consistent resting-state networks across healthy subjects. ***Proc Natl Acad Sci USA, 103*****, 13848-53.**

Dancause, N., Barbay, S., Frost, S. B., Plautz, E. J., Chen, D., Zoubina, E. V., et al. (2005). Extensive cortical rewiring after brain injury. ***The Journal of neuroscience : the official journal of the Society for Neuroscience, 25*****(44), 10167-79.**

de Haan, W., Mott, K., van Straaten, E. C., Scheltens, P., & Stam, C. J. (2012). Activity dependent degeneration explains hub vulnerability in Alzheimer's disease. ***PLoS computational biology, 8*****(8), e1002582.**

de Pasquale, F., Della Penna, S., Snyder, A. Z., Lewis, C., Mantini, D., Marzetti, L., et al. (2010). Temporal dynamics of spontaneous MEG activity in brain networks. ***Proceedings of the National Academy of Sciences of the United States of America, 107*****(13), 6040-5.**

Deco, G., Tononi, G., Boly, M., & Kringelbach, M. L. (2015). Rethinking segregation and integration: contributions of whole-brain modelling. ***Nature reviews Neuroscience, 16*****(7), 430-9.**

Di Pino, G., Pellegrino, G., Assenza, G., Capone, F., Ferreri, F., Formica, D., et al. (2014). Modulation of brain plasticity in stroke: a novel model for neurorehabilitation. ***Nature reviews Neurology, 10*****(10), 597-608.**

Dimyan, M. A., & Cohen, L. G. (2011). Neuroplasticity in the context of motor rehabilitation after stroke. *Nature reviews Neurology, 7*(2), 76-85.

Dirnagl, U. (2012). Pathobiology of injury after stroke: the neurovascular unit and beyond. *Annals of the New York Academy of Sciences, 1268*, 21-5.

Dirnagl, U., & Endres, M. (2014). Found in translation: preclinical stroke research predicts human pathophysiology, clinical phenotypes, and therapeutic outcomes. *Stroke; a journal of cerebral circulation, 45*(5), 1510-8.

Dirnagl, U., Iadecola, C., & Moskowitz, M. A. (1999). Pathobiology of ischaemic stroke: an integrated view. *Trends in neurosciences, 22*(9), 391-7.

Dobkin, J. A., Levine, R. L., Lagreze, H. L., Dulli, D. A., Nickles, R. J., & Rowe, B. R. (1989). Evidence for transhemispheric diaschisis in unilateral stroke. *Archives of neurology, 46*(12), 1333-6.

Dronkers, N. F., Plaisant, O., Iba-Zizen, M. T., & Cabanis, E. A. (2007). Paul Broca's historic cases: high resolution MR imaging of the brains of Leborgne and Lelong. *Brain : a journal of neurology, 130*(Pt 5), 1432-41.

Ekman, M., Derrfuss, J., Tittgemeyer, M., & Fiebach, C. J. (2012). Predicting errors from reconfiguration patterns in human brain networks. *Proceedings of the National Academy of Sciences of the United States of America, 109*(41), 16714-9.

Endres, M., Engelhardt, B., Koistinaho, J., Lindvall, O., Meairs, S., Mohr, J. P., et al. (2008). Improving outcome after stroke: overcoming the translational roadblock. *Cerebrovascular diseases, 25*(3), 268-78.

Endres, M., Laufs, U., Liao, J. K., & Moskowitz, M. A. (2004). Targeting eNOS for stroke protection. *Trends in neurosciences, 27*(5), 283-9.

Engel, A. K., Fries, P., & Singer, W. (2001). Dynamic predictions: oscillations and synchrony in top-down processing. *Nature reviews Neuroscience, 2*(10), 704-16.

Feeney, D. M., & Baron, J. C. (1986). Diaschisis. ***Stroke; a journal of cerebral circulation, 17*****(5), 817-30.**

Filippini, N., MacIntosh, B. J., Hough, M. G., Goodwin, G. M., Frisoni, G. B., Smith, S. M., et al. (2009). Distinct patterns of brain activity in young carriers of the APOE-epsilon4 allele. ***Proceedings of the National Academy of Sciences of the United States of America, 106*****(17), 7209-14.**

Finger, S., Koehler, P. J., & Jagella, C. (2004). The Monakow concept of diaschisis: origins and perspectives. ***Archives of neurology, 61*****(2), 283-8.**

Fischl, B., Rajendran, N., Busa, E., Augustinack, J., Hinds, O., Yeo, B. T., et al. (2008). Cortical folding patterns and predicting cytoarchitecture. ***Cerebral cortex, 18*****(8), 1973-80.**

Fornito, A., Harrison, B. J., Zalesky, A., & Simons, J. S. (2012). Competitive and cooperative dynamics of large-scale brain functional networks supporting recollection. ***Proceedings of the National Academy of Sciences of the United States of America, 109*****(31), 12788-93.**

Fox, M. D., & Greicius, M. (2010). Clinical applications of resting state functional connectivity. ***Frontiers in systems neuroscience, 4*****, 19.**

Fox, M. D., & Raichle, M. E. (2007). Spontaneous fluctuations in brain activity observed with functional magnetic resonance imaging. ***Nat Rev Neurosci, 8*****(9), 700-11.**

Fox, M. D., Snyder, A. Z., Vincent, J. L., Corbetta, M., Van Essen, D. C., & Raichle, M. E. (2005). The human brain is intrinsically organized into dynamic, anticorrelated functional networks. ***Proceedings of the National Academy of Sciences of the United States of America, 102*****(27), 9673-8.**

Fox, M. D., Snyder, A. Z., Vincent, J. L., & Raichle, M. E. (2007). Intrinsic fluctuations within cortical systems account for intertrial variability in human behavior. ***Neuron, 56*****, 171-84.**

Fox, M. D., Snyder, A. Z., Zacks, J. M., & Raichle, M. E. (2006). Coherent spontaneous activity accounts for trial-to-trial variability in human evoked brain responses. ***Nature Neuroscience, 9*****(1), 23-5.**

Fox, P. T., & Friston, K. J. (2012). Distributed processing; distributed functions? *NeuroImage, 61*(2), 407-26.

Friston, K., & Kiebel, S. (2009). Predictive coding under the free-energy principle. *Philosophical transactions of the Royal Society of London Series B, Biological sciences, 364*(1521), 1211-21.

Friston, K. J. (1994). Functional and Effective Connectivity in Neuroimaging: A Synthesis. *Human brain mapping, 2*, 56-78.

Friston, K. J. (2011). Functional and effective connectivity: a review. *Brain connectivity, 1*(1), 13-36.

Friston, K. J., Homes, A., Worsley, K., Poline, J., Frith, C., & Frackowiak, R. S. (1994). Statistical parametric maps in functional imaging: A general linear approach. *Human Brain Mapping, 2*, 189-210.

Gillebert, C. R., & Mantini, D. (2013). Functional connectivity in the normal and injured brain. *The Neuroscientist : a review journal bringing neurobiology, neurology and psychiatry, 19*(5), 509-22.

Gillebert, C. R., Mantini, D., Thijs, V., Sunaert, S., Dupont, P., & Vandenberghe, R. (2011). Lesion evidence for the critical role of the intraparietal sulcus in spatial attention. *Brain : a journal of neurology, 134*(Pt 6), 1694-709.

Golestani, A. M., Tymchuk, S., Demchuk, A., & Goodyear, B. G. (2013). Longitudinal evaluation of resting-state FMRI after acute stroke with hemiparesis. *Neurorehabilitation and neural repair, 27*(2), 153-63.

Golland, Y., Bentin, S., Gelbard, H., Benjamini, Y., Heller, R., Nir, Y., et al. (2007). Extrinsic and intrinsic systems in the posterior cortex of the human brain revealed during natural sensory stimulation. *Cereb Cortex, 17*, 766-77.

Gratton, C., Nomura, E. M., Perez, F., & D'Esposito, M. (2012). Focal brain lesions to critical locations cause widespread disruption of the modular organization of the brain. *Journal of cognitive neuroscience, 24*(6), 1275-85.

Grefkes, C., & Fink, G. R. (2011). Reorganization of cerebral networks after stroke: new insights from neuroimaging with connectivity approaches. *Brain : a journal of neurology, 134*(Pt 5), 1264-76.

Grefkes, C., & Fink, G. R. (2012). Disruption of motor network connectivity post-stroke and its noninvasive neuromodulation. *Current opinion in neurology, 25*(6), 670-5.

Grefkes, C., Nowak, D. A., Eickhoff, S. B., Dafotakis, M., Kust, J., Karbe, H., et al. (2008). Cortical connectivity after subcortical stroke assessed with functional magnetic resonance imaging. *Annals of neurology, 63*(2), 236-46.

Grefkes, C., Nowak, D. A., Wang, L. E., Dafotakis, M., Eickhoff, S. B., & Fink, G. R. (2010). Modulating cortical connectivity in stroke patients by rTMS assessed with fMRI and dynamic causal modeling. *NeuroImage, 50*(1), 233-42.

Grefkes, C., & Ward, N. S. (2014). Cortical reorganization after stroke: how much and how functional? *The Neuroscientist : a review journal bringing neurobiology, neurology and psychiatry, 20*(1), 56-70.

Greicius, M. (2008). Resting-state functional connectivity in neuropsychiatric disorders. *Current opinion in neurology, 21*(4), 424-30.

Greicius, M. D., Flores, B. H., Menon, V., Glover, G. H., Solvason, H. B., Kenna, H., et al. (2007). Resting-state functional connectivity in major depression: abnormally increased contributions from subgenual cingulate cortex and thalamus. *Biol Psychiatry, 62*(5), 429-37.

Greicius, M. D., Kiviniemi, V., Tervonen, O., Vainionpaa, V., Alahuhta, S., Reiss, A. L., et al. (2008). Persistent default-mode network connectivity during light sedation. *Hum Brain Mapp, 29*(7), 839-47.

Greicius, M. D., Srivastava, G., Reiss, A. L., & Menon, V. (2004). Default-mode network activity distinguishes Alzheimer's disease from healthy aging: evidence from functional MRI. *Proc Natl Acad Sci U S A, 101*(13), 4637-42.

Greicius, M. D., Supekar, K., Menon, V., & Dougherty, R. F. (2009). Resting-state functional connectivity reflects structural connectivity in the default mode network. *Cerebral cortex, 19*(1), 72-8.

Grinvald, A., Arieli, A., Tsodyks, M., & Kenet, T. (2003). Neuronal assemblies: single cortical neurons are obedient members of a huge orchestra. ***Biopolymers, 68*****(3), 422-36.**

Hagmann, P., Cammoun, L., Gigandet, X., Meuli, R., Honey, C. J., Wedeen, V. J., et al. (2008). Mapping the structural core of human cerebral cortex. ***PLoS biology, 6*****(7), e159.**

Handwerker, D. A., Roopchansingh, V., Gonzalez-Castillo, J., & Bandettini, P. A. (2012). Periodic changes in fMRI connectivity. ***NeuroImage, 63*****(3), 1712-9.**

Hasson, U., Malach, R., & Heeger, D. J. (2010). Reliability of cortical activity during natural stimulation. ***Trends in cognitive sciences, 14*****(1), 40-8.**

He, B. J., Shulman, G. L., Snyder, A. Z., & Corbetta, M. (2007). The role of impaired neuronal communication in neurological disorders. ***Curr Opin Neurol, 20*****(6), 655-60.**

He, B. J., Snyder, A. Z., Vincent, J. L., Epstein, A., Shulman, G. L., & Corbetta, M. (2007). Breakdown of functional connectivity in frontoparietal networks underlies behavioral deficits in spatial neglect. ***Neuron, 53*****(6), 905-18.**

Hesselmann, G., Kell, C. A., & Kleinschmidt, A. (2008). Ongoing activity fluctuations in hMT+ bias the perception of coherent visual motion. ***Journal of Neuroscience, 28*****, 14481-5.**

Hesselmann, G., Kell, C.A., Eger, E., Kleinschmidt, A. (2008). Spontaneous local variations in ongoing neural activity bias perceptual decisions. ***PNAS, 105*****, 10984-9.**

Hipp, J. F., Hawellek, D. J., Corbetta, M., Siegel, M., & Engel, A. K. (2012). Large-scale cortical correlation structure of spontaneous oscillatory activity. ***Nature neuroscience, 15*****(6), 884-90.**

Honey, C. J., Kotter, R., Breakspear, M., & Sporns, O. (2007). Network structure of cerebral cortex shapes functional connectivity on multiple time scales. ***Proc Natl Acad Sci U S A, 104*****(24), 10240-5.**

Honey, C. J., & Sporns, O. (2008). Dynamical consequences of lesions in cortical networks. ***Human brain mapping, 29*****(7), 802-9.**

Honey, C. J., Sporns, O., Cammoun, L., Gigandet, X., Thiran, J. P., Meuli, R., et al. (2009). Predicting human resting-state functional connectivity from structural connectivity. ***Proceedings of the National Academy of Sciences of the United States of America, 106*****(6), 2035-40.**

Horn, A., Ostwald, D., Reisert, M., & Blankenburg, F. (2014). The structural-functional connectome and the default mode network of the human brain. ***NeuroImage, 102 Pt 1*****, 142-51.**

Hummel, F. C., & Cohen, L. G. (2006). Non-invasive brain stimulation: a new strategy to improve neurorehabilitation after stroke? ***The Lancet Neurology, 5*****(8), 708-12.**

Hutchison, R. M., Womelsdorf, T., Allen, E. A., Bandettini, P. A., Calhoun, V. D., Corbetta, M., et al. (2013). Dynamic functional connectivity: promise, issues, and interpretations. ***NeuroImage, 80*****, 360-78.**

Hutchison, R. M., Womelsdorf, T., Gati, J. S., Everling, S., & Menon, R. S. (2013). Resting-state networks show dynamic functional connectivity in awake humans and anesthetized macaques. ***Human brain mapping, 34*****(9), 2154-77.**

Joel, S. E., Caffo, B. S., van Zijl, P. C., & Pekar, J. J. (2011). On the relationship between seed-based and ICA-based measures of functional connectivity. ***Magnetic resonance in medicine, 66*****(3), 644-57.**

Johansen-Berg, H., Scholz, J., & Stagg, C. J. (2010). Relevance of structural brain connectivity to learning and recovery from stroke. ***Frontiers in systems neuroscience, 4*****, 146.**

Johansson, B. B. (2004). Brain plasticity in health and disease. ***The Keio journal of medicine, 53*****(4), 231-46.**

Johnston, M. V. (2004). Clinical disorders of brain plasticity. ***Brain & development, 26*****(2), 73-80.**

Jones, D. T., Vemuri, P., Murphy, M. C., Gunter, J. L., Senjem, M. L., Machulda, M. M., et al. (2012). Non-stationarity in the "resting brain's" modular architecture. ***PLoS ONE, 7*****(6), e39731.**

Keilholz, S. D., Magnuson, M. E., Pan, W. J., Willis, M., & Thompson, G. J. (2013). Dynamic properties of functional connectivity in the rodent. *Brain connectivity, 3*(1), 31-40.

Kelly, A. M., Margulies, D. S., & Castellanos, F. X. (2007). Recent advances in structural and functional brain imaging studies of attention-deficit/hyperactivity disorder. *Current Psychiatry Reports, 9*(5), 401-7.

Kenet, T., Bibitchkov, D., Tsodyks, M., Grinvald, A., & Arieli, A. (2003). Spontaneously emerging cortical representations of visual attributes. *Nature, 425*(6961), 954-6.

Kennedy, D. P., & Courchesne, E. (2008). The intrinsic functional organization of the brain is altered in autism. *Neuroimage, 39*(4), 1877-85.

Kinnison, J., Padmala, S., Choi, J. M., & Pessoa, L. (2012). Network analysis reveals increased integration during emotional and motivational processing. *The Journal of neuroscience : the official journal of the Society for Neuroscience, 32*(24), 8361-72.

Kiran, S. (2012). What is the nature of poststroke language recovery and reorganization? *ISRN neurology, 2012*, 786872.

Kiviniemi, V., Kantola, J. H., Jauhiainen, J., Hyvarinen, A., & Tervonen, O. (2003). Independent component analysis of nondeterministic fMRI signal sources. *NeuroImage, 19*(2 Pt 1), 253-60.

Ko, S. B., & Yoon, B. W. (2013). Mechanisms of functional recovery after stroke. *Frontiers of neurology and neuroscience, 32*, 1-8.

Kolb, B., & Gibb, R. (2014). Searching for the principles of brain plasticity and behavior. *Cortex; a journal devoted to the study of the nervous system and behavior, 58*, 251-60.

Kolb, B., & Whishaw, I. Q. (1998). Brain plasticity and behavior. *Annual review of psychology, 49*, 43-64.

Krainik, A., Hund-Georgiadis, M., Zysset, S., & von Cramon, D. Y. (2005). Regional impairment of cerebrovascular reactivity and BOLD signal in adults after stroke. ***Stroke; a journal of cerebral circulation, 36*****(6), 1146-52.**

Kunz, A., Dirnagl, U., & Mergenthaler, P. (2010). Acute pathophysiological processes after ischaemic and traumatic brain injury. ***Best practice & research Clinical anaesthesiology, 24*****(4), 495-509.**

Laird, A. R., Fox, P. M., Eickhoff, S. B., Turner, J. A., Ray, K. L., McKay, D. R., et al. (2011). Behavioral interpretations of intrinsic connectivity networks. ***Journal of cognitive neuroscience, 23*****(12), 4022-37.**

Lange, N., Strother, S. C., Anderson, J. R., Nielsen, F. A., Holmes, A. P., Kolenda, T., et al. (1999). Plurality and resemblance in fMRI data analysis. ***NeuroImage, 10*****(3 Pt 1), 282-303.**

Lassalle-Lagadec, S., Sibon, I., Dilharreguy, B., Renou, P., Fleury, O., & Allard, M. (2012). Subacute default mode network dysfunction in the prediction of post-stroke depression severity. ***Radiology, 264*****(1), 218-24.**

Laufs, H., & et al. (2003). Electroencephalographic signatures of attentional and cognitive default modes in spontaneous brain activity fluctuations at rest. ***Proc Natl Acad Sci USA, 100*****, 11053-8.**

Laufs, H., Holt, J. L., Elfont, R., Krams, M., Paul, J. S., Krakow, K., et al. (2006). Where the BOLD signal goes when alpha EEG leaves. ***Neuroimage, 31*****(4), 1408-18.**

Laufs, H., Kleinschmidt, A., Beyerle, A., Eger, E., Salek-Haddadi, A., Preibisch, C., et al. (2003). EEG-correlated fMRI of human alpha activity. ***Neuroimage, 19*****(4), 1463-76.**

Leopold, D. A., Murayama, Y., & Logothetis, N. K. (2003). Very slow activity fluctuations in monkey visual cortex: implications for functional brain imaging. ***Cereb Cortex, 13*****(4), 422-33.**

Llinas, R. R. (1988). The intrinsic electrophysiological properties of mammalian neurons: insights into central nervous system function. ***Science, 242*****(4886), 1654-64.**

Logothetis, N. K. (2003). The underpinnings of the BOLD functional magnetic resonance imaging signal. ***J Neurosci, 23*****(10), 3963-71.**

Lohmann, G., Ovadia-Caro, S., Jungehulsing, G. J., Margulies, D. S., Villringer, A., & Turner, R. (2012). Connectivity concordance mapping: a new tool for model-free analysis of FMRI data of the human brain. ***Frontiers in systems neuroscience, 6*****, 13.**

Lowe, M. J., Mock, B. J., & Sorenson, J. A. (1998). Functional connectivity in single and multislice echoplanar imaging using resting-state fluctuations. ***Neuroimage, 7*****(2), 119-32.**

Lv, Y., Margulies, D. S., Cameron Craddock, R., Long, X., Winter, B., Gierhake, D., et al. (2013). Identifying the perfusion deficit in acute stroke with resting-state functional magnetic resonance imaging. ***Annals of neurology, 73*****(1), 136-40.**

Mantini, D., Perrucci, M. G., Del Gratta, C., Romani, G. L., & Corbetta, M. (2007). Electrophysiological signatures of resting state networks in the human brain. ***Proc Natl Acad Sci U S A, 104*****(32), 13170-5.**

Margulies, D. S., Bottger, J., Long, X., Lv, Y., Kelly, C., Schafer, A., et al. (2010). Resting developments: a review of fMRI post-processing methodologies for spontaneous brain activity. ***Magma, 23*****(5-6), 289-307.**

Markowitsch, H. J., & Pritzel, M. (1978). Von Monakow's diaschisis concept: comments on West et al. (1976). ***Behavioral biology, 22*****(3), 411-2.**

Mazzetto-Betti, K. C., Leoni, R. F., Pontes-Neto, O. M., Santos, A. C., Leite, J. P., Silva, A. C., et al. (2010). The stability of the blood oxygenation level-dependent functional MRI response to motor tasks is altered in patients with chronic ischemic stroke. ***Stroke; a journal of cerebral circulation, 41*****(9), 1921-6.**

McAvoy, M., Larson-Prior, L., Nolan, T. S., Vaishnavi, S. N., Raichle, M. E., & d'Avossa, G. (2008). Resting states affect spontaneous BOLD oscillations in sensory and paralimbic cortex. ***Journal of neurophysiology, 100*****(2), 922-31.**

Meinzer, M., Darkow, R., Lindenberg, R., & Floel, A. (2016). Electrical stimulation of the motor cortex enhances treatment outcome in post-stroke aphasia. *Brain : a journal of neurology, 139*(Pt 4), 1152-63.

Milgram, S. (1967). The small world problem. *Psychol Today, 1*, 61-7.

Miller, J. E., Ayzenshtat, I., Carrillo-Reid, L., & Yuste, R. (2014). Visual stimuli recruit intrinsically generated cortical ensembles. *Proceedings of the National Academy of Sciences of the United States of America, 111*(38), E4053-61.

Moosmann, M., Ritter, P., Krastel, I., Brink, A., Thees, S., Blankenburg, F., et al. (2003). Correlates of alpha rhythm in functional magnetic resonance imaging and near infrared spectroscopy. *NeuroImage, 20*(1), 145-58.

Mozaffarian, D., Benjamin, E. J., Go, A. S., Arnett, D. K., Blaha, M. J., Cushman, M., et al. (2015). Heart disease and stroke statistics--2015 update: a report from the American Heart Association. *Circulation, 131*(4), e29-322.

Mueller, S., Wang, D., Fox, M. D., Yeo, B. T., Sepulcre, J., Sabuncu, M. R., et al. (2013). Individual variability in functional connectivity architecture of the human brain. *Neuron, 77*(3), 586-95.

Murphy, K., Birn, R. M., & Bandettini, P. A. (2013). Resting-state fMRI confounds and cleanup. *NeuroImage, 80*, 349-59.

Murphy, T. H., & Corbett, D. (2009). Plasticity during stroke recovery: from synapse to behaviour. *Nature reviews Neuroscience, 10*(12), 861-72.

Napieralski, J. A., Butler, A. K., & Chesselet, M. F. (1996). Anatomical and functional evidence for lesion-specific sprouting of corticostriatal input in the adult rat. *The Journal of comparative neurology, 373*(4), 484-97.

Nir, Y., Hasson, U., Levy, I., Yeshurun, Y., & Malach, R. (2006). Widespread functional connectivity and fMRI fluctuations in human visual cortex in the absence of visual stimulation. *Neuroimage, 30*(4), 1313-24.

Nir, Y., Mukamel, R., Dinstein, I., Privman, E., Harel, M., Fisch, L., et al. (2008). Interhemispheric correlations of slow spontaneous neuronal fluctuations revealed in human sensory cortex. ***Nature Neuroscience, 11*****, 1100-8.**

Nomura, E. M., Gratton, C., Visser, R. M., Kayser, A., Perez, F., & D'Esposito, M. (2010). Double dissociation of two cognitive control networks in patients with focal brain lesions. ***Proceedings of the National Academy of Sciences of the United States of America, 107*****(26), 12017-22.**

Nudo, R. J. (2013). Recovery after brain injury: mechanisms and principles. ***Frontiers in human neuroscience, 7*****, 887.**

Opitz, A., Fox, M. D., Craddock, R. C., Colcombe, S., & Milham, M. P. (2016). An integrated framework for targeting functional networks via transcranial magnetic stimulation. ***NeuroImage, 127*****, 86-96.**

Ovadia-Caro, S., Margulies, D. S., & Villringer, A. (2014). The value of resting-state functional magnetic resonance imaging in stroke. ***Stroke; a journal of cerebral circulation, 45*****(9), 2818-24.**

Ovadia-Caro, S., Nir, Y., Soddu, A., Ramot, M., Hesselmann, G., Vanhaudenhuyse, A., et al. (2012). Reduction in inter-hemispheric connectivity in disorders of consciousness. ***PLoS ONE, 7*****(5), e37238.**

Ovadia-Caro, S., Villringer, K., Fiebach, J., Jungehulsing, G. J., van der Meer, E., Margulies, D. S., et al. (2013). Longitudinal effects of lesions on functional networks after stroke. ***Journal of cerebral blood flow and metabolism : official journal of the International Society of Cerebral Blood Flow and Metabolism*****.**

Overman, J. J., & Carmichael, S. T. (2014). Plasticity in the injured brain: more than molecules matter. ***The Neuroscientist : a review journal bringing neurobiology, neurology and psychiatry, 20*****(1), 15-28.**

Park, C. H., Chang, W. H., Ohn, S. H., Kim, S. T., Bang, O. Y., Pascual-Leone, A., et al. (2011). Longitudinal changes of resting-state functional connectivity during motor recovery after stroke. ***Stroke; a journal of cerebral circulation, 42*****(5), 1357-62.**

Pascual-Leone, A., Amedi, A., Fregni, F., & Merabet, L. B. (2005). The plastic human brain cortex. ***Annual review of neuroscience, 28*****, 377-401.**

Pineiro, R., Pendlebury, S., Johansen-Berg, H., & Matthews, P. M. (2002). Altered hemodynamic responses in patients after subcortical stroke measured by functional MRI. ***Stroke; a journal of cerebral circulation, 33*****(1), 103-9.**

Power, J. D., Barnes, K. A., Snyder, A. Z., Schlaggar, B. L., & Petersen, S. E. (2012). Spurious but systematic correlations in functional connectivity MRI networks arise from subject motion. ***NeuroImage, 59*****(3), 2142-54.**

Raichle, M. E. (2006). Neuroscience. The brain's dark energy. ***Science, 314*****(5803), 1249-50.**

Raichle, M. E. (2009). A paradigm shift in functional brain imaging. ***J Neurosci, 29*****(41), 12729-34.**

Raichle, M. E. (2015). The restless brain: how intrinsic activity organizes brain function. ***Philosophical transactions of the Royal Society of London Series B, Biological sciences, 370*****(1668).**

Raichle, M. E., & Mintun, M. A. (2006). Brain work and brain imaging. ***Annual Review of Neuroscience, 29*****, 449-76.**

Redecker, C., Luhmann, H. J., Hagemann, G., Fritschy, J. M., & Witte, O. W. (2000). Differential downregulation of GABAA receptor subunits in widespread brain regions in the freeze-lesion model of focal cortical malformations. ***The Journal of neuroscience : the official journal of the Society for Neuroscience, 20*****(13), 5045-53.**

Rehme, A. K., Eickhoff, S. B., Rottschy, C., Fink, G. R., & Grefkes, C. (2012). Activation likelihood estimation meta-analysis of motor-related neural activity after stroke. ***NeuroImage, 59*****(3), 2771-82.**

Rehme, A. K., Fink, G. R., von Cramon, D. Y., & Grefkes, C. (2011). The role of the contralesional motor cortex for motor recovery in the early days after stroke assessed with longitudinal FMRI. ***Cerebral cortex, 21*****(4), 756-68.**

Reijneveld, J. C., Ponten, S. C., Berendse, H. W., & Stam, C. J. (2007). The application of graph theoretical analysis to complex networks in the brain. ***Clinical neurophysiology : official journal of the International Federation of Clinical Neurophysiology, 118*****(11), 2317-31.**

Rombouts, S. A., Barkhof, F., Goekoop, R., Stam, C. J., & Scheltens, P. (2005). Altered resting state networks in mild cognitive impairment and mild Alzheimer's disease: an fMRI study. *Hum Brain Mapp, 26*(4), 231-9.

Rossini, P. M., Calautti, C., Pauri, F., & Baron, J. C. (2003). Post-stroke plastic reorganisation in the adult brain. *Lancet Neurol, 2*(8), 493-502.

Sacco, R. L., Kasner, S. E., Broderick, J. P., Caplan, L. R., Connors, J. J., Culebras, A., et al. (2013). An updated definition of stroke for the 21st century: a statement for healthcare professionals from the American Heart Association/American Stroke Association. *Stroke; a journal of cerebral circulation, 44*(7), 2064-89.

Saur, D., & Hartwigsen, G. (2012). Neurobiology of language recovery after stroke: lessons from neuroimaging studies. *Archives of Physical Medicine and Rehabilitation, 93*(1 Suppl), S15-25.

Saur, D., Lange, R., Baumgaertner, A., Schraknepper, V., Willmes, K., Rijntjes, M., et al. (2006). Dynamics of language reorganization after stroke. *Brain : a journal of neurology, 129*(Pt 6), 1371-84.

Saur, D., Ronneberger, O., Kummerer, D., Mader, I., Weiller, C., & Kloppel, S. (2010). Early functional magnetic resonance imaging activations predict language outcome after stroke. *Brain : a journal of neurology, 133*(Pt 4), 1252-64.

Schaefer, A., Margulies, D. S., Lohmann, G., Gorgolewski, K. J., Smallwood, J., Kiebel, S. J., et al. (2014). Dynamic network participation of functional connectivity hubs assessed by resting-state fMRI. *Frontiers in human neuroscience, 8*, 195.

Schaefer, A., Quinque, E., Kipping, J., Arélin, K., Frisch, S., Roggenhofer, E., et al. (2014). Early small vessel disease affects frontoparietal and cerebellar hubs in close correlation with clinical symptoms – A resting-state fMRI study. *Journal of Cerebral Blood Flow and Metabolism, In press*.

Sehm, B., Schafer, A., Kipping, J., Margulies, D., Conde, V., Taubert, M., et al. (2012). Dynamic modulation of intrinsic functional connectivity by transcranial direct current stimulation. *Journal of neurophysiology, 108*(12), 3253-63.

Shehzad, Z., Kelly, A. M., Reiss, P. T., Gee, D. G., Gotimer, K., Uddin, L. Q., et al. (2009). The resting brain: unconstrained yet reliable. ***Cereb Cortex, 19*****(10), 2209-29.**

Skudlarski, P., Jagannathan, K., Calhoun, V. D., Hampson, M., Skudlarska, B. A., & Pearlson, G. (2008). Measuring brain connectivity: diffusion tensor imaging validates resting state temporal correlations. ***NeuroImage, 43*****(3), 554-61.**

Smith, S. M., Fox, P. T., Miller, K. L., Glahn, D. C., Fox, P. M., Mackay, C. E., et al. (2009). Correspondence of the brain's functional architecture during activation and rest. ***Proceedings of the National Academy of Sciences of the United States of America, 106*****(31), 13040-5.**

Sporns, O. (2011). From simple graphs to the connectome: Networks in neuroimaging. ***NeuroImage*****.**

Sporns, O. (2013). Network attributes for segregation and integration in the human brain. ***Current opinion in neurobiology, 23*****(2), 162-71.**

Sporns, O., Chialvo, D. R., Kaiser, M., & Hilgetag, C. C. (2004). Organization, development and function of complex brain networks. ***Trends in cognitive sciences, 8*****(9), 418-25.**

Sporns, O., Tononi, G., & Kotter, R. (2005). The human connectome: A structural description of the human brain. ***PLoS computational biology, 1*****(4), e42.**

Stam, C. J., & Reijneveld, J. C. (2007). Graph theoretical analysis of complex networks in the brain. ***Nonlinear biomedical physics, 1*****(1), 3.**

Stark, D. E., Margulies, D. S., Shehzad, Z. E., Reiss, P., Kelly, A. M., Uddin, L. Q., et al. (2008). Regional variation in interhemispheric coordination of intrinsic hemodynamic fluctuations. ***The Journal of neuroscience : the official journal of the Society for Neuroscience, 28*****(51), 13754-64.**

Striem-Amit, E., Ovadia-Caro, S., Caramazza, A., Margulies, D. S., Villringer, A., & Amedi, A. (2015). Functional connectivity of visual cortex in the blind follows retinotopic organization principles. ***Brain : a journal of neurology, 138*****(Pt 6), 1679-95.**

Tombari, D., Loubinoux, I., Pariente, J., Gerdelat, A., Albucher, J. F., Tardy, J., et al. (2004). A longitudinal fMRI study: in recovering and then in clinically stable sub-cortical stroke patients. ***NeuroImage, 23*****(3), 827-39.**

Tononi, G., Sporns, O., & Edelman, G. M. (1994). A measure for brain complexity: relating functional segregation and integration in the nervous system. ***Proceedings of the National Academy of Sciences of the United States of America, 91*****(11), 5033-7.**

Tuladhar, A. M., Snaphaan, L., Shumskaya, E., Rijpkema, M., Fernandez, G., Norris, D. G., et al. (2013). Default Mode Network Connectivity in Stroke Patients. ***PLoS ONE, 8*****(6).**

Tzourio-Mazoyer, N., Landeau, B., Papathanassiou, D., Crivello, F., Etard, O., Delcroix, N., et al. (2002). Automated anatomical labeling of activations in SPM using a macroscopic anatomical parcellation of the MNI MRI single-subject brain. ***NeuroImage, 15*****(1), 273-89.**

Urban, E. T., 3rd, Bury, S. D., Barbay, H. S., Guggenmos, D. J., Dong, Y., & Nudo, R. J. (2012). Gene expression changes of interconnected spared cortical neurons 7 days after ischemic infarct of the primary motor cortex in the rat. ***Molecular and cellular biochemistry, 369*****(1-2), 267-86.**

van de Ven, V. G., Formisano, E., Prvulovic, D., Roeder, C. H., & Linden, D. E. (2004). Functional connectivity as revealed by spatial independent component analysis of fMRI measurements during rest. ***Human brain mapping, 22*****(3), 165-78.**

van den Heuvel, M., Mandl, R., Luigjes, J., & Hulshoff Pol, H. (2008). Microstructural organization of the cingulum tract and the level of default mode functional connectivity. ***The Journal of neuroscience : the official journal of the Society for Neuroscience, 28*****(43), 10844-51.**

van den Heuvel, M. P., Mandl, R. C., Kahn, R. S., & Hulshoff Pol, H. E. (2009). Functionally linked resting-state networks reflect the underlying structural connectivity architecture of the human brain. ***Human brain mapping, 30*****(10), 3127-41.**

Van Dijk, K. R., Hedden, T., Venkataraman, A., Evans, K. C., Lazar, S. W., & Buckner, R. L. (2010). Intrinsic functional connectivity as a tool for human connectomics: theory, properties, and optimization. ***Journal of neurophysiology, 103*****(1), 297-321.**

van Meer, M. P., Otte, W. M., van der Marel, K., Nijboer, C. H., Kavelaars, A., van der Sprenkel, J. W., et al. (2012). Extent of bilateral neuronal network reorganization and functional recovery in relation to stroke severity. ***The Journal of neuroscience : the official journal of the Society for Neuroscience, 32*****(13), 4495-507.**

van Meer, M. P., van der Marel, K., Otte, W. M., Berkelbach van der Sprenkel, J. W., & Dijkhuizen, R. M. (2010). Correspondence between altered functional and structural connectivity in the contralesional sensorimotor cortex after unilateral stroke in rats: a combined resting-state functional MRI and manganese-enhanced MRI study. ***Journal of cerebral blood flow and metabolism : official journal of the International Society of Cerebral Blood Flow and Metabolism, 30*****(10), 1707-11.**

van Meer, M. P., van der Marel, K., Wang, K., Otte, W. M., El Bouazati, S., Roeling, T. A., et al. (2010). Recovery of sensorimotor function after experimental stroke correlates with restoration of resting-state interhemispheric functional connectivity. ***The Journal of neuroscience : the official journal of the Society for Neuroscience, 30*****(11), 3964-72.**

Vanhaudenhuyse, A., Noirhomme, Q., Tshibanda, L. J., Bruno, M. A., Boveroux, P., Schnakers, C., et al. (2010). Default network connectivity reflects the level of consciousness in non-communicative brain-damaged patients. ***Brain, 133*****(Pt 1), 161-71.**

Villringer, A., & Dirnagl, U. (1995). Coupling of brain activity and cerebral blood flow: basis of functional neuroimaging. ***Cerebrovascular and brain metabolism reviews, 7*****(3), 240-76.**

Vincent, J. L., Patel, G. H., Fox, M. D., Snyder, A. Z., Baker, J. T., van Essen, D. C., et al. (2007). Intrinsic functional architecture in the anaesthetized monkey brain. ***Nature, 447*****, 83-8.**

von Monakow, C. (1914). Die Lokalisation im Grosshirn und der Abbau der Funktion durch kortikale Herde. ***Wiesbaden: JF Bergmann***

von Monakow, C. (1969). Diaschisis [1914 article translated by G Harris], in Pribram KH (ed): *Brain and Behavior I: Moods, States and Mind* (pp. 27-36). Baltimore: Penguin.

Wang, K., Liang, M., Wang, L., Tian, L., Zhang, X., Li, K., et al. (2007). Altered functional connectivity in early Alzheimer's disease: a resting-state fMRI study. *Hum Brain Mapp, 28*(10), 967-78.

Wang, L., Yu, C., Chen, H., Qin, W., He, Y., Fan, F., et al. (2010). Dynamic functional reorganization of the motor execution network after stroke. *Brain : a journal of neurology, 133*(Pt 4), 1224-38.

Ward, N. S. (2005). Plasticity and the functional reorganization of the human brain. *International journal of psychophysiology : official journal of the International Organization of Psychophysiology, 58*(2-3), 158-61.

Ward, N. S., Brown, M. M., Thompson, A. J., & Frackowiak, R. S. (2003). Neural correlates of motor recovery after stroke: a longitudinal fMRI study. *Brain : a journal of neurology, 126*(Pt 11), 2476-96.

Ward, N. S., & Cohen, L. G. (2004). Mechanisms underlying recovery of motor function after stroke. *Archives of neurology, 61*(12), 1844-8.

Watts, D. J., & Strogatz, S. H. (1998). Collective dynamics of 'small world' networks. *Nature, 393*, 440-2.

Weissenbacher, A., Kasess, C., Gerstl, F., Lanzenberger, R., Moser, E., & Windischberger, C. (2009). Correlations and anticorrelations in resting-state functional connectivity MRI: a quantitative comparison of preprocessing strategies. *NeuroImage, 47*(4), 1408-16.

West, J. R. (1978). The concept of diaschisis: a reply to Markowitsch and Pritzel. *Behavioral biology, 22*(3), 413-6.

West, J. R., Deadwyler, S. A., Cotman, C. W., & Lynch, G. S. (1976). An experimental test of diaschisis. *Behavioral biology, 18*(3), 419-25.

Yuste, R., MacLean, J. N., Smith, J., & Lansner, A. (2005). The cortex as a central pattern generator. *Nature reviews Neuroscience, 6*(6), 477-83.

Zhou, Y., Liang, M., Tian, L., Wang, K., Hao, Y., Liu, H., et al. (2007). Functional disintegration in paranoid schizophrenia using resting-state fMRI. ***Schizophr Res, 97*****(1-3), 194-205.**

7 Supplements

7.1 curriculum vitae

Name	Smadar A. Ovadia-Caro
Date of Birth	14.12.1979
Place of Birth	Tel-Aviv, Israel
Nationality	Israeli
Address	Humboldt-Universitāt zu Berlin Berlin School of Mind and Brain Luisenstraße 56, 10099 Berlin Tel. +49 (0) 30 2093-6034 Email: Smadar.ovadia@gmail.com

EDUCATION

2010- Present	**Department of psychology and Berlin School of Mind and Brain, Humboldt University**, Berlin. **Max Planck Institute for Human Cognitive and Brain Sciences**, Leipzig. Doctoral student Advisors: Prof. Villringer A and Prof. van der Meer E.
2007- 2009	**Weizmann Institute of Science,** Feinberg Graduate School, Rehovot, Israel. M.Sc. program, Department of Neurobiology Thesis advisor: Prof. Rafael Malach Thesis: *Decreased inter-hemispheric correlation in patients with disorders of consciousness: an fMRI study.* M.Sc., November 2009.
2007- 2009	**Haifa University**, Haifa, Israel. Faculty of Social Welfare and Health Sciences. B.P.T., June 2006, Summa Cum Laude.

SCHOLARSHIPS AND ACADEMIC AWARDS

2010	Doctoral fellowship, Berlin School of Mind and Brain, Humboldt University, Berlin.
2010	Doctoral Fellowship, Minerva Foundation, Max-Planck Society.

2010	Exchange fellowship, Daniel Turnberg Trust Fund. A visiting student at the lab of Prof. Owen A., Cambridge UK.
2009	Travel award, Berlin School of Mind and Brain.
2002-2006	Excellence scholarship (four consecutive scholarships based on academic achievements).

TEACHING

2011	Medical neuroscience program, Charité – Universitätsmedizin Berlin, Germany. Co-lecturer with Dr. Daniel Margulies Lecture title: Blood Oxygen-Level Dependent (BOLD) Functional Magnetic Resonance Imaging: General introduction, learning-induced changes in the healthy brain, and applications to assessment of post-stroke rehabilitation.
2010	Seconda Università di Napoli, Italy. Introduction to BrainVoyager (neuroimaging software), course developer and instructor on the behalf of the Malach Lab, Weizmann Institute of Science, Israel.
2005-2006	Student union, Haifa University, Israel. Tutor. Academic assistance for students with socio-economical difficulties or those of minority groups.

INVITED TALKS

2013	5th International Stroke Symposium, Berlin, Germany. Title: *Resting-state fMRI and Stroke*

POSTER PRESENTATIONS

Longitudinal effects of lesions on functional networks after stroke. Organization for Human Brain Mapping. Seattle, USA, June 16 - 20, 2013.

Dynamics of functional connectivity following lacunar stroke. The 3rd Biennial Conference on Resting-state Brain Connectivity. Magdeburg, Germany, September 5 – 7, 2012.

The effects of congenitally blindness in humans on intrinsic functional brain systems: a functional magnetic resonance imaging study. International Society for Eye Research. Berlin, Germany, July 21 – 25, 2012.

Increased functional connectivity between middle frontal gyrus and striatum in pathological gambling. Organization for Human Brain Mapping. Beijing, China, June 10 – 14, 2012.

Decreased inter-hemispheric correlation in patients with disorders of consciousness. Berlin Brain Days. Berlin, Germany, December 9 – 11, 2009.

AD HOC REVIEWER

Journal of Cerebral Blood Flow and Metabolism
Frontiers in Human Neuroscience
Neuroimage Clinical

7.2 publication list

Striem-Amit E.*, **Ovadia-Caro S.*,** Caramazza A., Margulies DS., Villringer A., Amedi A. *Functional connectivity of visual cortex in the blind follows retinotopic organization principles*. Brain, 2015. 138 (6). PMID: 25869851. *** Equal contribution**.

Ovadia-Caro S., Margulies DS., Villringer A. *The value of resting-state functional magnetic resonance imaging in stroke*. Stroke, 2014. 45 (9). PMID: 25013022.

Ovadia-Caro S., Villringer K., Jan Jungehulsing G., Van der Meer E., Margulies D.S., Villringer A. *Longitudinal effects of lesions on functional networks after stroke.* Journal of Cerebral Blood Flow and Metabolism, 2013. 33(8). PMID: 23715061.

Köhler S., **Ovadia-Caro S.,** Villringer A., Heinz A., Seiferth N., Margulies D.S. *Increased functional connectivity between prefrontal cortex and reward system in pathological gambling*. PloS ONE, 2013. 8(12):e84565. PMID: 24367675.

Ovadia-Caro S., Nir Y., Soddu A., Ramot M., Hesselmann G., Vanhaudenhuyse A., Dinstein I., Tshibanda JF-L., Boly M., Harel M., Laureys S., and Malach R. *Reduction in inter-hemispheric connectivity in disorders of consciousness.* PloS ONE, 2012. 7(5): e37238. PMID: 22629375.

Lohmann G., **Ovadia-Caro S.,** Jan Jungehulsing G., Margulies D.S., Villringer A., Turner A*. Connectivity concordance mapping: a new tool for model-free analysis of fMRI data of the human brain*. Frontiers in system neuroscience, 2012. 6 (13). PMID: 22470320.

Soddu, A., Vanhaudenhuyse, A., Bahri, M. A., Bruno, M-A., Boly, M., Demertzi, A., Tshibanda, J.-F., Phillips, C., Stanziano, M., **Ovadia-Caro, S**., Nir, Y., Maquet, P., Papa, M., Malach, R., Laureys, S. and Noirhomme, Q. *Identifying the default-mode component in spatial IC analyses of patients with disorders of consciousness*. Hum. Brain Mapp, 2011. 33(4). PMID: 21484953.

Soddu A, Boly M, Nir Y, Noirhomme Q, Vanhaudenhuyse A, Demertzi A, Arzi A, **Ovadia S,** Stanziano M, Papa M, Laureys S, Malach R. *Reaching across the abyss: recent advances in functional magnetic resonance imaging and their potential relevance to disorders of consciousness*. Progress in Brain Research, 2009. 177. PMID: 19818907.

7.3 research articles

frontiers in
SYSTEMS NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 20 March 2012
doi: 10.3389/fnsys.2012.00013

Connectivity concordance mapping: a new tool for model-free analysis of fMRI data of the human brain

Gabriele Lohmann[1]*, Smadar Ovadia-Caro[2], Gerhard Jan Jungehülsing[3], Daniel S. Margulies[1,2], Arno Villringer[1,2,3] and Robert Turner[1]

[1] *Max Planck Institute for Human Cognitive and Brain Sciences, Leipzig, Germany*
[2] *Mind and Brain Institute and Berlin School of Mind and Brain, Berlin, Germany*
[3] *Center for Stroke Research, Charité – Universitätsmedizin, Berlin, Germany*

Edited by:
Per E. Roland, Karolinska Institute, Sweden

Reviewed by:
Stefano Panzeri, Italian Institute of Technology, Italy
Anders Ledberg, Universitat Pompeu Fabra, Spain

****Correspondence:***
Gabriele Lohmann, Max Planck Institute for Human Cognitive and Brain Sciences, Stephanstr. 1a, 04103 Leipzig, Germany.
e-mail: lohmann@cbs.mpg.de

Functional magnetic resonance data acquired in a task-absent condition ("resting state") require new data analysis techniques that do not depend on an activation model. Here, we propose a new analysis method called Connectivity Concordance Mapping (CCM). The main idea is to assign a label to each voxel based on the reproducibility of its whole-brain pattern of connectivity. Specifically, we compute the correlations of time courses of each voxel with every other voxel for each measurement. Voxels whose correlation pattern is consistent across measurements receive high values. The result of a CCM analysis is thus a voxel-wise map of concordance values. Regions of high inter-subject concordance can be assumed to be functionally consistent, and may thus be of specific interest for further analysis. Here we present two fMRI studies to demonstrate the possible applications of the algorithm. The first is a eyes-open/eyes-closed paradigm designed to highlight the potential of the method in a relatively simple domain. The second study is a longitudinal repeated measurement of a patient following stroke. Longitudinal clinical studies such as this may represent the most interesting domain of applications for this algorithm.

Keywords: connectivity, resting state

INTRODUCTION

In recent years, spontaneous blood oxygen level dependent (BOLD) fluctuations have gained interest in the field of neuroscience. The popularity of intrinsic brain activity, often operationalized as "resting state" functional magnetic resonance imaging (fMRI), is in large part due to its use in mapping functional connectivity. Such connectivity maps are based on the intrinsic correlation between distributed brain regions. They have been shown to replicate the spatial extent of task-related functional systems without needing an overt task (e.g., Biswal et al., 1995), and are highly reproducible across individuals (Damoiseaux et al., 2006), time (Shehzad et al., 2009), and sites (Biswal et al., 2010). These features make intrinsic functional connectivity a particularly promising approach to describing functional brain organization across various populations and brain states. However, truly data-driven exploratory analytic techniques are still needed in the field.

The growing interest in functional connectivity is closely related to resting state fMRI (rs-fMRI). One of the most important points in favor of rs-fMRI is its attractiveness for clinical applications (Fox and Greicius, 2010). In patients, using resting state fMRI is advantageous mainly due to the lack of task involvement. Some patients are unable to perform certain tasks, or rather have different performance level, which could influence the results. In addition, rs-fMRI is relatively easy to carry out and is less time consuming, which makes it ideal for the clinical population.

As a result, there is an increased demand for simple analysis methods that require little manual intervention. Such methods cannot draw on information of any experimental design and should be as parameter-free as possible. The earliest, and most commonly used analysis of spontaneous BOLD fluctuations involves defining a region-of-interest, or "seed region," and correlating it with all other voxels in the brain. Although the "seed-based" functional connectivity is intuitive and straightforward in its results, the main disadvantage is the requirement of an *a priori* hypothesis as to the location of the seed (Biswal et al., 1995; Fox et al., 2006; Margulies et al., 2007). In contrast, independent component analysis (ICA) has gained wide popularity as a data-driven technique that distinguishes functional brain networks (Beckmann et al., 2005; Damoiseaux et al., 2006; De Luca et al., 2006; Smith et al., 2009). Nonetheless, *a posteriori* decisions are required to determine which components are functionally relevant. (For reviews of issues relating to these methodologies, see: Cole et al., 2010; Margulies et al., 2010.) These are both limiting factors for an analysis that allows discovery-based investigation of functional brain organization (Biswal et al., 2010).

Graph-based measures have gained interest recently (Bullmore and Sporns, 2009; Buckner, 2010; Sporns, 2010). Some of these approaches are computationally very complex and require a reduction in the number of "nodes" in the brain through the use of anatomically (Tzourio-Mazoyer et al., 2002) and functionally defined (Craddock et al., 2011) parcelation units. Centrality-based methods (Buckner, 2010; Lohmann et al., 2010) detect "hubs" as special features of networks, but may fail to find other relevant features.

Here, we propose a new method of fMRI data analysis called "Connectivity Concordance Mapping (CCM)." The aim of CCM is to express the reproducibility of functional connectivity patterns in the whole brain. Specifically, we compute the correlations of time courses of each voxel with every other voxel in each dataset. Voxels whose correlation pattern is consistent across datasets receive high values. Consistency is measured using Kendall's W (Kendall and Babington Smith, 1939). The result of a CCM analysis is a voxel-wise map of concordance values. Regions of high concordance can be assumed to be functionally consistent, and may thus be of specific interest for further analysis, such as a seed-based analysis. Concordance can be measured between subjects (i.e., inter-subject consistency), or in the same subject between different time points (i.e., longitudinal dataset). Here we present an example for each one of these scenarios. Longitudinal studies are of particular interest in clinical environments where it is often important to monitor the progression of a disease, or the effectiveness of a treatment.

In the context of fMRI experiments, concordance has been used to assess reproducibility (e.g., Lange et al., 1999; Shehzad et al., 2009; Biswal et al., 2010) or regional homogeneity (Zang et al., 2004; Liu et al., 2010, see also Hasson et al., 2009) for a review about reproducibility of activation patterns. The novelty of the present approach is that concordance can be used as a tool for whole-brain mapping at the voxel level. Our method is closely related to another approach recently proposed by Shehzad et al. (2011) called MDMR. MDMR calculates the main effect of how much patterns of connectivity are predicted by a phenotypic and possibly continuous variable. In contrast, we are primarily interested in assessing concordance in connectivity between groups of subjects or across longitudinal repeated measurements. Another difference is that we propose to use Kendall's W as a concordance measure whereas Shezad et al. use linear correlation. As will be discussed in the following section, for the types of application we have in mind here, concordance is often better suited than correlation.

MATERIALS AND METHODS

ALGORITHM

We define a region-of-interest (ROI) containing n voxels to which all subsequent analysis steps are restricted. In the experiments reported in this study, the ROIs covered the entire cerebrum and consisted of $n \approx 50{,}000$ voxels.

We assume that m fMRI data sets exist and are geometrically aligned to some common geometrical reference frame. These data sets may for instance be acquired from different subjects participating in the same study or from one subject undergoing repeated measurements in a longitudinal study. We are interested in the concordance of these data sets with respect to their connectivity pattern.

For each data set $k = 1,\ldots, m$ and each voxel address $i = 1,\ldots, n$ we obtain a connectivity vector $\mathbf{s_i^k} = (s^k_{i,1}, \ldots, s^k_{i,n})$ whose entries $s^k_{i,j}$ contain a pairwise similarity measure between the time courses of voxels i and j. Several similarity measures may be used for this purpose. In this study, we use Pearson's linear correlation coefficient defined as:

$$r_{xy} = \frac{\sum_t (x_t - \bar{x})(y_t - \bar{y})}{\sqrt{\sum_t (x_t - \bar{x})^2 (y_t - \bar{y})^2}} \tag{1}$$

with $x_t, y_t, t = 1,\ldots, T$ time series in two voxels, and their $\bar{x}, \bar{y}$ temporal means. Other similarity metrics such as wavelet coherence or spectral coherence may be used as well. Here, we are interested in the concordance of the connectivity vectors $\mathbf{s_i^k}$ across the multiple measurements $k = 1,\ldots, m$. This concordance value is used to label voxel i.

Several concordance measures are known, and are discussed in a large body of literature (e.g., Shrout and Fleiss, 1979; Lin, 1989; Barnhart et al., 2002). Here, we propose to use Kendall's W (Kendall and Babington Smith, 1939; Legendre, 2005) as a measure of concordance. It yields values between 0 and 1 with "1" denoting complete agreement and "0" complete disagreement. Its main advantage over other concordance measures is its ease of use and the fact that it does not presuppose Gaussianity of the data. This is particularly important when connectivity measures other than linear correlation are used.

Kendall's W is defined as follows.

Let $\mathbf{s_k^i}$, $k = 1,\ldots, m$ be m connectivity vectors in voxel i as defined above. Suppose for instance, that the m different data sets correspond to m different "raters" who are asked to rank each voxel j according to the strength of its correlation with the current voxel i. As a first step, we perform a ranking such that r_{jk} denotes the j-th rank among the n entries of vector $\mathbf{s_k^i}$. The total rank given to each voxel j by those raters is

$$R_j = \sum_{k=1}^{m} r_{jk}, \quad j = 1, \ldots, n \tag{2}$$

The mean values of these total ranks is

$$\bar{R} = 0.5 * m * (n + 1) \tag{3}$$

and the sum of squared deviations is

$$S = \sum_{j=1}^{n} \left(R_j - \bar{R}\right)^2. \tag{4}$$

If the fictitious raters disagree, their ranking should be approximately random so that the sum of their ranks is about equal and their deviation S low, while a high value of S indicates good agreement. Thus, with some normalization terms, Kendall's W can be defined as

$$W = \frac{12S}{m^2(n^3 - n)}. \tag{5}$$

For comparison, we also tested the "Overall Concordance Correlation Coefficient (OCCC)" (Barnhart et al., 2002) defined as

$$\rho = \frac{2 \sum_{j=1}^{m-1} \sum_{k=j+1}^{m} C_{jk}}{(m-1) \sum_{j=1}^{m} C_j^2 + m \sum_{j=1}^{m} (s_{\cdot j} - s_{\cdot\cdot})^2} \tag{6}$$

with s_j, $j = 1,\ldots, m$ observation vectors which in our case correspond to the correlation maps described above, and $s_{\cdot j}$, $s_{\cdot\cdot}$, C_j^2 and C_{jk} as sample means, overall mean, variances and covariances, respectively. Note that in contrast to Kendall's W, OCCC

presupposes Gaussianity of the data. For further detail see Barnhart et al. (2002).

One should note that concordance differs from correlation in several aspects. Most importantly, linear correlation is based on a subtraction of the mean. Therefore, when using correlation instead of concordance one could imagine two connectivity maps that show the same pattern, where in one map correlations are generally much lower. In such a situation, using Pearson's correlation the consistency between the two maps would be rated high whereas Kendall's W would give a low mark. In many applications, one would like to distinguish those two cases. In clinical applications for instance, it may be important to monitor global changes in connectivity which would not be possible using correlation instead of concordance.

EXPERIMENT 1

As an illustration of the CCM pattern during "resting state" condition in healthy controls, we applied CCM to two resting state paradigms: eyes-open and eyes-closed. Functional magnetic resonance imaging (fMRI) data were obtained in a 30.4 min (800 volumes) resting state scan using a Siemens Tim Trio 3T scanner at the University Hospital Charite in Berlin, Germany for 7 healthy subjects. Three-dimensional functional images using blood oxygen level dependent (BOLD) contrast were obtained with a gradient echo planar imaging (EPI) sequence (TR = 2300 ms, TE = 30 ms, 30 slices; voxel size: 3.125 mm × 3.125 mm × 4 mm, flip angle 90°). The scan was comprised of four blocks of 7.6 min each. The experiment was approved by the local ethics committee. In two of the four blocks subjects were instructed to keep their eyes-open, in the other two blocks eyes were closed, with no overt task being imposed. No fixation cross was presented. The order of the two conditions was randomized across subjects. For the present study, we analyzed the first eyes-open/eyes-closed block (each condition lasting 200 volume). Data were preprocessed using the software package Lipsia (Lohmann et al., 2001). Preprocessing of functional data included slice-scan time and motion correction, alignment to a functional template of MNI space (resampling to an isotropic voxel grid with a resolution of 3 mm^3), band pass filtering (0.011–0.166 Hz) and spatial smoothing of 7 mm FWHM. Global signal was not removed.

For the purpose of CCM analysis a region-of-interest was manually defined. The mask contained 47,872 voxels covering the entire cerebrum (see **Figure A1** in Appendix). CCM was computed in two ways: (1) inter-subject CCM for each one of the conditions separately (i.e. eyes-open/eyes-closed). Each condition was computed based on time series from the 7 subjects. (2) Intra-subject CCM was computed for each subject's eyes-closed and eyes-open scan and was later averaged across subjects (n = 7).

EXPERIMENT 2

As an illustration of the potential usefulness of CCM in clinical data, we applied CCM to a longitudinal data set acquired in a patient following lacunar stroke. fMRI data were obtained in a 5.75 min (150 volumes) resting state scan using a Siemens Tim Trio 3T scanner at the University Hospital Charite in Berlin, Germany. The patient was instructed to lie still and keep the eyes-open for the duration of the scan, with no overt task being imposed. The patient was scanned at four time points: day 1, day 27, day 94, and day 199 following the stroke (corresponding to t1, t2, t3, and t4; day 0 represents the day of the stroke). Three-dimensional functional images using BOLD contrast were obtained with a EPI sequence (TR = 2300 ms, TE = 30 ms, 30 slices; voxel size: 3.125 mm × 3.125 mm × 4 mm, flip angle 90°). T1-weighted anatomical images were acquired using a 3D MPRAGE sequence (TR = 1900 ms, TE = 2.52 ms, TI = 900 ms, 192 slices, voxel size: 1 mm × 1 mm × 1 mm, flip angle 9°). Diffusion Weighted imaging was acquired in t1 for the purpose of lesion localization. Data were preprocessed using the software package Lipsia (Lohmann et al., 2001). Preprocessing included slice-scan time and motion correction, alignment to an anatomical MNI template based on the T1-MPRAGE image acquired at t3 (resampling to an isotropic voxel grid with a resolution of 3 mm^3), band pass filtering (0.0125–0.166 Hz) and spatial smoothing of 6 mm FWHM. Global signal was not removed. For the purpose of CCM analysis a region-of-interest was manually defined. The mask contained 50,199 voxels covering the entire cerebrum and parts of the cerebellum. Ventricles were manually excluded from the mask (see **Figure A2** in Appendix). An independent radiologist determined stroke location. Stroke was localized to left thalamus and left occipital cortex due to posterior cerebral artery infarct (see **Figure 4**). The ethics committee of the Faculty of Medicine at the Charite University, Berlin, Germany, approved the experiment. Written informed consent was obtained from the patient. CCM was computed for all four time points (i.e., t1–t4) and for pairs of consecutive time points (i.e., t1 and t2, t2 and t3, t3 and t4), in order to trace changes in the connectivity pattern over shorter time points as well. As a second level of analysis, linear correlation based on a seed was computed for each one of the time points. A seed voxel was defined in the left thalamus based on a local maximum of CCM value. A comparison between two time points was done by a subtraction of the corresponding functional connectivity maps.

RESULTS

EXPERIMENT 1

In order to explore the typical pattern of CCM during common resting state paradigms, CCM was computed for 7 subjects in the two resting states of experiment 1. **Figure 1** depicts the CCM inter-subjects' results using Kendall's W as a concordance measure. In both states, most of the cortical gray matter is concordant including the thalamus subcortically. Areas that demonstrate high concordance include the visual, auditory and sensory-motor networks. Additionally, areas which are known to be part of the task negative network, also known as the "default mode network" (Gusnard et al., 2001), such as posterior cingulate cortex, inferior parietal lobule and middle frontal gyrus are concordant but to a lesser extent than primary regions regions. Lateral areas of the prefrontal cortex are less concordant than the rest of the brain. For comparison, **Figure 2** shows the same data analyzed using the Overall Concordance Correlation Coefficient (OCCC; Barnhart et al., 2002). Note that the results appear quite similar. The Pearson correlation between the maps of **Figure 1** compared to **Figure 2** was 0.94 for the eyes-closed condition, and 0.91 for the eyes-open condition.

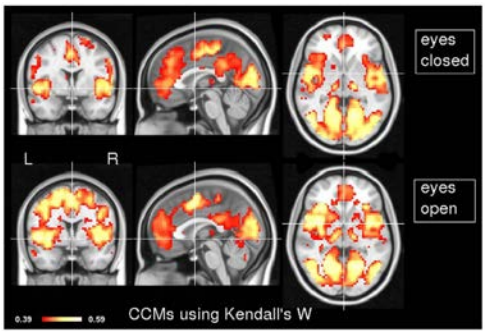

FIGURE 1 | Inter-subject CCM using Kendall's W. CCM has been calculated for eyes-closed (top) and eyes-open (bottom), n = 7 subjects in each condition. MNI coordinates of slice positions are (0, 0, 0). The concordance measure was Kendall's W.

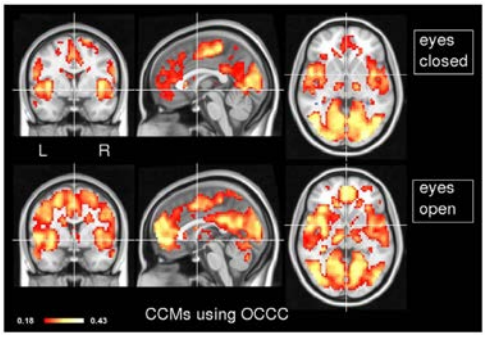

FIGURE 2 | Inter-subject CCM using OCCC. As in **Figure 1**, CCM has been calculated for eyes-closed (top) and eyes-open (bottom), n = 7 subjects in each condition. MNI coordinates of slice positions are (0, 0, 0). For comparison, the concordance measure was OCCC.

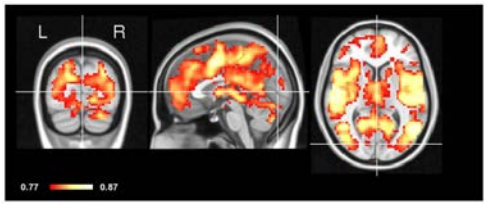

FIGURE 3 | Intra-subject CCM eyes-open and eyes-closed. The concordance of eyes-open and eyes-closed scans was assessed within each subject separately yielding an individual CCM per subject. These CCMs were then averaged. Note that early visual cortex (V1) shows relatively low concordance compared to other brain regions. Crosshair is positioned in the Calcarine sulcus.

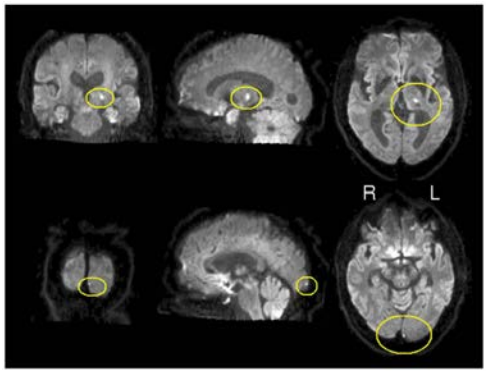

FIGURE 4 | Location of stroke. Diffusion weighted image. Hyper intensities marked in yellow represent the lesioned areas. Lesions are restricted to left thalamus and left occipital pole.

In order to test the concordance between the two states, we applied CCM to the eyes-open and eyes-closed datasets in the same subject (within-subject CCM) and computed an average of the resulting maps. As can be seen in **Figure 3**, most of the areas of high concordance are similar to the spatial pattern of CCM in the inter-subject CCM, with the exception of primary visual cortex (V1) which is notably smaller in concordance.

EXPERIMENT 2

In order to explore the potential applicability of CCM to clinical data sets, concordance was measured in a patient following stroke in a longitudinal manner. **Figure 4** depicts the location of the stroke on a diffusion weighted image acquired one day after stroke. The lesion was restricted to left thalamus and an additional lesion in the left occipital cortex. As a first step of the analysis, we applied CCM on all four time points following the stroke; day 1, day 27, day 94, and day 199 following the stroke (corresponding to t1, t2, t3, and t4, day 0 represents the day of the stroke). **Figure 5** shows concordance across all time points (t1, t2, t3, t4). As in the healthy subjects of experiment 1, cortical regions were generally found to be concordant in this patient. For investigating the process of recovery after stroke, we monitored changes in connectivity represented by low concordance. The lowest concordance value of the entire brain was found in the left globus pallidus with Kendall's W = 0.22 (**Figure 5C**).

In order to assess dynamics of concordance over time, we computed CCMs in each pair of consecutive time points i.e., t1 and t2, t2 and t3, and t3 and t4 (**Figure 6**). As before, most of the gray matter showed high concordance values. However, some areas show marked changes in their concordance over time (**Figure 7**). Prominent changes in the left and right globus pallidus were found in the difference between the CCMs of t2, t3 versus t3, t4, but much less in the difference of the CCMs of t1, t2 versus t2, t3. This indicates

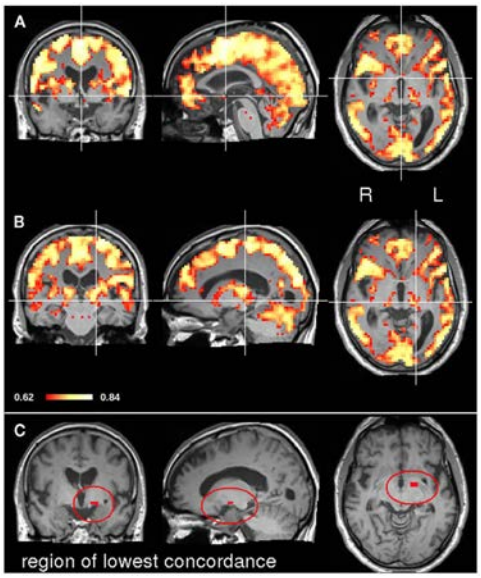

FIGURE 5 | CCM in a stroke patient. The concordance over all four time points (t1, t2, t3, and t4) was assessed. The top image **(A)** shows the CCM map at MNI slice position (0, 0, 0). In the middle image **(B)** the map crosshair marks the position of the left thalamic lesion. The bottom image **(C)** shows the region where connectivity concordance was lowest.

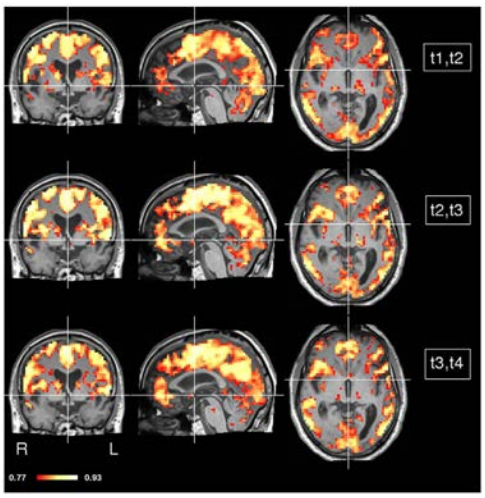

FIGURE 6 | CCM dynamics in a stroke patient. The concordance between any two consecutive time points was calculated (t1 and t2, t2 and t3, t3 and t4). MNI coordinates of slice positions are (0, 0, 0).

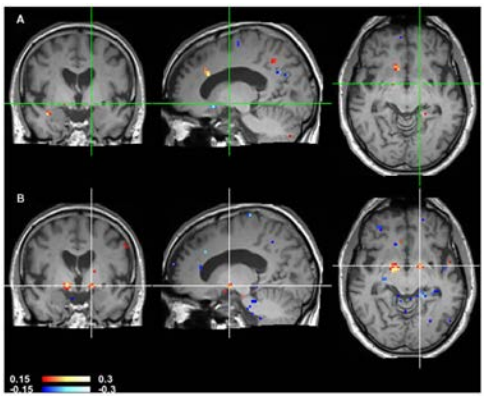

FIGURE 7 | CCM dynamics in a stroke patient. CCMs of consecutive time points of **Figure 6** were subtracted to highlight changes over time. The top image **(A)** shows CCM(t1, t2) minus CCM(t2, t3). The bottom image **(B)** shows CCM(t2, t3) minus CCM(t3, t4). Note that globus pallidus marked by the crosshair shows a large difference in the second difference image but not in the first indicating that the changes in this area occur at a late stage.

that the low concordance that we found in the overall concordance map shown in **Figure 5** is primarily a late stage effect.

Local maxima or minima in connectivity concordance maps may be used as seeds for subsequent correlation analyses. Since the results shown in **Figure 6** indicated a change in connectivity with globus pallidus, we used the local minima located in the left and right globus pallidus as seed voxels (**Figures 8** and **9**). We found a noticeable change in inter-hemispheric differences between time points t3 and t4.

DISCUSSION

In this study, we have introduced a new algorithm called connectivity concordance mapping (CCM) for model-free analysis of fMRI data. CCM computes the voxel-wise concordance of fMRI time courses and can be applied to regions-of-interest as large as the whole brain. One should note that Kendall's W may be biased by underlying correlations within brain structures. As a result, it is possible that Kendall's W may differ between two brain regions, although the underlying correlational structure within each respective region remains identical. Therefore, CCM should be used only as a method for generating hypotheses to be further explored and validated in subsequent processing steps such as seed-based correlation analyses.

We implemented CCM on resting state data in healthy subjects in different functional states. Our main finding is that CCM can differentiate between different functional states. A comparison between Kendall's W and OCCC as measures of concordance did not show major differences. However, since OCCC presupposes Gaussianity of the data, we suggest to use Kendall's W because it is applicable to a wider range of cases.

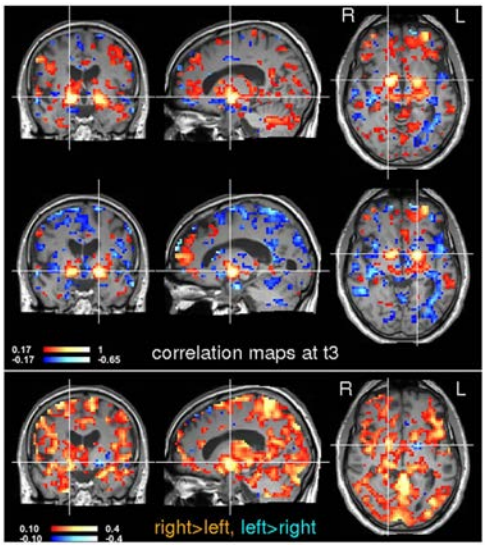

FIGURE 8 | Seed-based correlation analysis at time t3. Voxels in the left and right globus pallidus were used for a subsequent seed-based correlation analysis. The location of the seeds is indicated by the crosshairs. Inter-hemispheric differences are shown in the bottom image. Note that the right globus pallidus generally shows stronger correlations than the left.

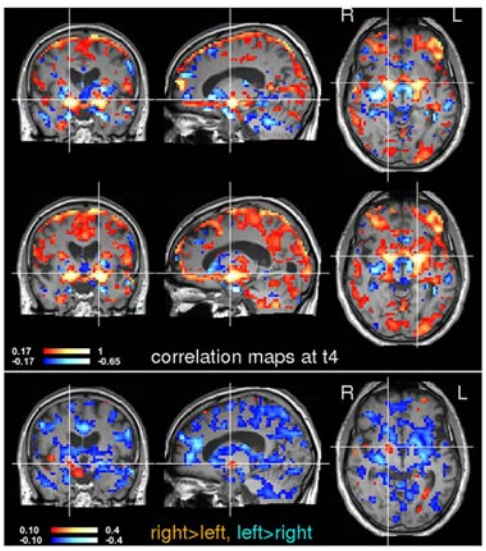

FIGURE 9 | Seed-based correlation analysis at time t4. Voxels in the left and right globus pallidus were used for a subsequent seed-based correlation analysis. The location of the seeds is indicated by the crosshairs. Note that inter-hemispheric differences have changed noticeably between t3 and t4 (see **Figure 8**).

The investigation of state-dependent (eyes-opened/closed) differences using CCM implicated primary visual cortex, revealing within-subject concordance contrasts throughout the cuneus. Such a finding offers strong support to the functional significance of CCM. Previous studies have investigated the impact of eyes-opened and eyes-closed states on resting state functional connectivity results (McAvoy et al., 2008; Van Dijk et al., 2010). Our current results independently replicated the findings of McAvoy et al. (2008), who initially demonstrated that the functional connectivity of the visual cortex was altered in the eyes-closed condition, and that its spectral density in low-frequencies was higher. Likewise, Van Dijk et al. (2010) found that the eyes-closed condition demonstrated diminished functional connectivity in the default mode and attention networks, when compared to eyes-opened or eyes-opened with fixation. While our findings are not novel contributions to the understanding of the network differences between these two task conditions, our aim was rather to demonstrate how the methodology employed here could detect these same differences without any *a priori* assumptions about what areas were involved. Such a voxel-wise approach provides a powerful exploratory tool for investigating more complex, and less well understood state-related differences.

One should note that inter-subject CCMs may be confounded by anatomical variability because concordance may be lower in areas of high anatomical variance. This is not a concern for longitudinal data as such the one reported in experiment 2 because here we used only one single subject (the stroke patient). In experiment 1 however, it may have influenced the results. Anatomical variability is a very general problem not restricted to CCM that affects all group studies including task-based designs analyzed by the standard general linear model (GLM). Inter-subject alignment procedures can alleviate (but not solve) the problem to some extent.

Longitudinal studies are one of the most relevant areas of application of the CCM approach and may be used to explore clinical data. In the exploratory CCM analysis of a patient following stroke, we found that CCM can be used to provide insight into the dynamics of functional connectivity changes over the course of recovery. It can also serve a basis for subsequent seed-based analysis. In the present case study, we found inter-hemispheric differences in correlation with left and right globus pallidus between times t3 and t4 (day 94 and day 199). This effect was picked up by CCM so that globus pallidus appeared as a local minimum in the CCM maps. Globus pallidus is part of the extrapyramidal motor system which was effected by the thalamic stroke in this patient. The CCM analysis suggests that recovery after stroke involved this system.

We should note however that this finding is quite preliminary. The purpose of the present experiment was to provide a "proof of concept" for the use of CCM as a tool for monitoring recovery after stroke as a test case for similar clinical applications. A more careful interpretation of our results will follow in subsequent

studies. In particular, since functional connectivity and behavior has been shown to correlate after stroke (He et al., 2007; Warren et al., 2009; Carter et al., 2010; Wang et al., 2010), in order to interpret the impact of our results on the recovery process, additional behavioral information must be included. For a detailed review on connectivity related changes following stroke see Grefkes and Fink (2011).

While further research is necessary to elucidate the significance of such results, the corroboration of similar findings using our exploratory methods supports further application of CCM to more complex state differences. Possible future applications may be clinically oriented, exploring dynamics of recovery as well as assessing impact of different treatment interventions on changes in functional connectivity.

ACKNOWLEDGMENTS

The authors would like to thank Dr. med. Kersten Villringer, University Clinic Charite, Berlin, Germany.

REFERENCES

Barnhart, H., Haber, M., and Song, J. (2002). Overall concordance correlation coefficient for evaluating agreement among multiple observers. *Biometrics* 58, 1020–1027.

Beckmann, C., DeLuca, M., Devlin, J., and Smith, S. (2005). Investigations into resting-state connectivity using independent component analysis. *Philos. Trans. R. Soc. Lond. B Biol. Sci.* 360, 1001–1013.

Biswal, B., Mennes, M., Zuo, X. N., Gohel, S., Kelly, C., Smith, S. M., Beckmann, C. F., Adelstein, J. S., Buckner, R. L., Colcombe, S., Dogonowski, A. M., Ernst, M., Fair, D., Hampson, M., Hoptman, M. J., Hyde, J. S., Kiviniemi, V. J., Kötter, R., Li, S. J., Lin, C. P., Lowe, M. J., Mackay, C., Madden, D. J., Madsen, K. H., Margulies, D. S., Mayberg, H. S., McMahon, K., Monk, C. S., Mostofsky, S. H., Nagel, B. J., Pekar, J. J., Peltier, S. J., Petersen, S. E., Riedl, V., Rombouts, S. A., Rypma, B., Schlaggar, B. L., Schmidt, S., Seidler, R. D., Siegle, G. J., Sorg, C., Teng, G. J., Veijola, J., Villringer, A., Walter, M., Wang, L., Weng, X. C., Whitfield-Gabrieli, S., Williamson, P., Windischberger, C., Zang, Y. F., Zhang, H. Y., Castellanos, F. X., and Milham, M. P. (2010). Toward discovery science of human brain function. *Proc. Natl. Acad. Sci. U.S.A.* 107, 4734–4739.

Biswal, B., Yetkin, F., Haughton, V., and Hyde, J. (1995). Functional connectivity in the motor cortex of resting human brain using echo-planar MRI. *Magn. Reson. Med.* 34, 537–541.

Buckner, R. (2010). Human functional connectivity: new tools, unresolved questions. *Proc. Natl. Acad. Sci. U.S.A.* 107, 10769–10770.

Bullmore, E., and Sporns, O. (2009). Complex brain networks: graph theoretical analysis of structural and functional systems. *Nat. Rev. Neurosci.* 10, 186–198.

Carter, A., Astafiev, S., Lang, C., Connor, L., Rengachary, J., Strube, M., Pope, D., Shulman, G., and Corbetta, M. (2010). Resting interhemispheric functional magnetic resonance imaging connectivity predicts performance after stroke. *Ann. Neurol.* 67, 365–375.

Cole, D., Smith, S., and Beckmann, C. (2010). Advances and pitfalls in the analysis and interpretation of resting-state fMRI data. *Front. Syst. Neurosci.* 4:8. doi:10.3389/fnsys.2010.00008

Craddock, R., James, G., Holtzheimer, P., Hu, X., and Mayberg, H. (2011). A whole brain fMRI atlas generated via spatially constrained spectral clustering. *Hum. Brain Mapp.* doi:10.1002/hbm.21333. [Epub ahead of print].

Damoiseaux, J., Rombouts, S., Barkhof, F., Scheltens, P., Stam, C., Smith, S., and Beckmann, C. (2006). Consistent resting-state networks across healthy subjects. *Proc. Natl. Acad. Sci. U.S.A.* 103, 13848–13853.

De Luca, M., Beckmann, C., De Stefano, N., Matthews, P., and Smith, S. (2006). fMRI resting state networks define distinct modes of long-distance interactions in the human brain. *Neuroimage* 29, 1359–1367.

Fox, M., Corbetta, M., Snyder, A., Vincent, J., and Raichle, M. (2006). Spontaneous neuronal activity distinguishes human dorsal and ventral attention systems. *Proc. Natl. Acad. Sci. U.S.A.* 103, 10046–10051.

Fox, M., and Greicius, M. (2010). Clinical applications of resting state functional connectivity. *Front. Syst. Neurosci.* 4:19. doi:10.3389/fnsys.2010.000192008

Grefkes, C., and Fink, G. (2011). Reorganization of cerebral networks after stroke: new insights from neuroimaging with connectivity approaches. *Brain* 134, 1264–1276.

Gusnard, D., Akbudak, E., Shulman, G., and Raichle, M. (2001). Medial prefrontal cortex and self-referential mental activity: relation to a default mode of brain function. *Proc. Natl. Acad. Sci. U.S.A.* 98, 4259–4264.

Hasson, U., Malach, R., and Heeger, D. (2009). Reliability of cortical activity during natural stimulation. *Trends Cogn. Sci. (Regul. Ed.)* 14, 40–47.

He, B., Snyder, A., Vincent, J., Epstein, A., Shulman, G., and Corbetta, M. (2007). Breakdown of functional connectivity in frontoparietal networks underlies behavioral deficits in spatial neglect. *Neuron* 53, 905–918.

Kendall, M., and Babington Smith, B. (1939). The problem of m rankings. *Ann. Math. Stat.* 10, 275–287.

Lange, N., Strother, S., Anderson, J., Nielsen, F., Holmes, A., Kolenda, T., Savoy, R., and Hansen, L. K. (1999). Plurality and resemblance in fMRI data analysis. *Neuroimage* 10, 282–303.

Legendre, P. (2005). Species associations: the Kendall coefficient of concordance revisited. *J. Agric. Biol. Environ. Stat.* 10, 226–245.

Lin, L. (1989). A concordance correlation coefficient to evaluate reproducibility. *Biometrics* 45, 255–268.

Liu, D., Yan, C., Ren, J., Yao, L., Kiviniemi, V., and Zang, Y. (2010). Using coherence to measure regional homogeneity of resting state fMRI signal. *Front. Syst. Neurosci.* 4:24. doi:10.3389/fnsys.2010.00024

Lohmann, G., Margulies, D., Horstmann, A., Pleger, B., Lepsien, J. Goldhahn, D., Schloegl, H., Stumvoll, M., Villringer, A., and Turner, R. (2010). Eigenvector centrality mapping for analyzing connectivity patterns in fMRI data of the human brain. *PLoS ONE* 5, e10232. doi:10.1371/journal.pone.0010232

Lohmann, G., Müller, K., Bosch, V., Mentzel, H., Hessler, S., Chen, L., Zysset, S., and von Cramon, D. Y. (2001). LIPSIA – a new software system for the evaluation of functional magnetic resonance images of the human brain. *Comput. Med. Imaging Graph.* 25, 449–457.

Margulies, D., Böttger, J., Long, X., Lv, Y., Kelly, C., Schäfer, A., Goldhahn, D., Abbushi, A., Milham, M., Lohmann, G., and Villringer, A. (2010). Resting developments: a review of fMRI post-processing methodologies for spontaneous brain activity. *MAGMA* 23, 289–307.

Margulies, D., Kelly, A., Uddin, L., Biswal, B., Castellanos, F., and Milham, M. (2007). Mapping the functional connectivity of anterior cingulate cortex. *Neuroimage* 37, 579–588.

McAvoy, M., Larson-Prior, L., Nolan, T., Vaishnavi, S., Raichle, M., and d'Avossa, G. (2008). Resting states affect spontaneous bold oscillations in sensory and paralimbic cortex. *J. Neurophysiol.* 100, 922–931.

Shehzad, Z., Kelly, C., Reiss, P., Gee, D., Gotimer, K., Uddin, L. Q., Lee, S. H., Margulies, D. S., Roy, A. K., Biswal, B. B., Petkova, E., Castellanos, F. X., Milham, M. P. (2009). The resting brain: unconstrained yet reliable. *Cereb. Cortex* 19, 2209–2229.

Shehzad, Z., Reiss, P., Adelstein, J., Emerson, J., Chabernaud, C., Mennes, M., Di Martino, A., Kelly, C., Castellanos, F., and Milham, M. (2011). "Connectome-wide association studies (CWAS): a multivariate distance-based approach," in *Annual Meeting of the Organization for Human Brain Mapping*, Quebec, CA.

Shrout, P., and Fleiss, J. (1979). Intraclass correlation: uses in assessing rater reliability. *Psychol. Bull.* 86, 420–428.

Smith, S., Fox, P., Miller, K., Glahn, D., Fox, P., Mackay, C., Filippini, N., Watkins, K., Toro, R., Laird, A., and Beckmann, C. (2009). Correspondence of the brain's functional architecture during activation and rest. *Proc. Natl. Acad. Sci. U.S.A.* 106, 13040–13045.

Sporns, O. (2010). *Networks of the Brain*. Cambridge, MA: MIT Press.

Tzourio-Mazoyer, N., Landeau, B., Papathanassiou, D., Crivello, F., Etard, O., Delcroix, N., Mazoyer, B., and Joliot, M. (2002). Automated anatomical labeling of activations in SPM using a macroscopic anatomical parcellation of the MNI MRI single-subject brain. *Neuroimage* 15, 273–289.

Van Dijk, K., Hedden, T., Venkataraman, A., Evans, K., Lazar, S., and

Buckner, R. (2010). Intrinsic functional connectivity as a tool for human connectomics: theory, properties, and optimization. *J. Neurophysiol.* 103, 297–321.

Wang, L., Yu, C., Chen, H., Qin, W., He, Y., Fan, F., Zhang, Y., Wang, M., Li, K., Zang, Y., Woodward, T., and Zhu, C. (2010). Dynamic functional reorganization of the motor execution network after stroke. *Brain* 133, 1224–1238.

Warren, J., Crinion, J., Lambon Ralph, M., and Wise, R. (2009). Anterior temporal lobe connectivity correlates with functional outcome after aphasic stroke. *Brain* 132, 3428–3442.

Zang, Y., Jiang, T., Lu, Y., He, Y., and Tian, L. (2004). Regional homogeneity approach to fMRI data analysis. *Neuroimage* 22, 394–400.

Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Received: 15 November 2011; paper pending published: 20 December 2011; accepted: 29 February 2012; published online: 20 March 2012.

Citation: Lohmann G, Ovadia-Caro S, Jungehülsing GJ, Margulies DS, Villringer A and Turner R (2012) Connectivity concordance mapping: a new tool for model-free analysis of fMRI data of the human brain. Front. Syst. Neurosci. ***6****:13. doi: 10.3389/fnsys.2012.00013*

APPENDIX

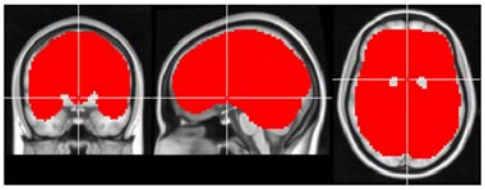

FIGURE A1 | Region-of-interest used for CCM analysis in experiment 1. The crosshair is located in MNI coordinates (0, 0, 0).

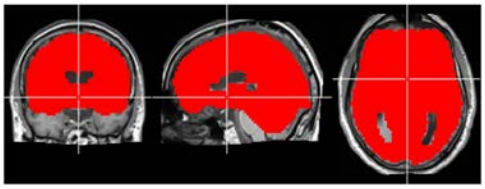

FIGURE A2 | Region-of-interest used for CCM analysis in experiment 2. The crosshair is located in MNI coordinates (0, 0, 0).

Journal of Cerebral Blood Flow & Metabolism (2013) 33, 1279–1285

www.jcbfm.com

ORIGINAL ARTICLE

Longitudinal effects of lesions on functional networks after stroke

Smadar Ovadia-Caro[1,2,3], Kersten Villringer[4], Jochen Fiebach[4], Gerhard Jan Jungehulsing[4,5], Elke van der Meer[1,3], Daniel S Margulies[1,2] and Arno Villringer[1,2,4]

While ischemic stroke reflects focal damage determined by the affected vascular territory, clinical symptoms are often more complex and may be better explained by additional indirect effects of the focal lesion. Assumed to be structurally underpinned by anatomical connections, supporting evidence has been found using alterations in the functional connectivity of resting-state functional magnetic resonance imaging (fMRI) data in both sensorimotor and attention networks. To assess the generalizability of this phenomenon in a stroke population with heterogeneous lesions, we investigated the distal effects of lesions on a global level. Longitudinal resting-state fMRI scans were acquired at three consecutive time points, beginning during the acute phase (days 1, 7, and 90 post-stroke) in 12 patients after ischemic stroke. We found a preferential functional change in affected networks (i.e., networks containing lesions changed more during recovery when compared with unaffected networks). This change in connectivity was significantly correlated with clinical changes assessed with the National Institute of Health Stroke Scale. Our results provide evidence that the functional architecture of large-scale networks is critical to understanding the clinical effect and trajectory of post-stroke recovery.

Journal of Cerebral Blood Flow & Metabolism (2013) **33,** 1279–1285; doi:10.1038/jcbfm.2013.80; published online 29 May 2013

Keywords: concordance; dual regression; heterogeneous lesions; intrinsic functional connectivity; resting-state fMRI

INTRODUCTION

The brain is a complex network of interacting functionally and structurally connected regions. As brain functions emerge from interacting regions that are part of a network topology,[1–4] so too can alterations across networks coincide with various pathological states such as disorders of consciousness,[5,6] Alzheimer's disease,[7] neuropsychiatric disorders,[8] and stroke.[9–13] Stroke lesions provide a unique model of how local damage can result in long-distance alterations. Similar structural damage has been shown to result in different levels of impairment to functionality, and patients can present multiple deficits in the acute phase that are not easily attributed to the direct effect of the focal lesion.[10,14] The complex symptoms not explained by damage to the infarct core are partially explained by hypoperfusion in penumbral areas surrounding the lesion,[15,16] as well as effects on distal regions connected to the damaged tissue.[17]

Using functional connectivity measures based on temporal correlation of spontaneous blood oxygenation level-dependent (BOLD) signal fluctuations (resting-state fMRI), it has previously been shown that localized brain lesions can cause connectivity-based changes in regions that are structurally intact and far from the lesion site. This phenomenon has been demonstrated in the motor[13,18,19] and attention networks,[9,20] and has also been shown to correlate with behavioral improvement in the post-stroke recovery phase.[9,13,18–20]

Heterogeneity of lesions in terms of location and size is one of the notorious challenges in stroke research. A generalizable analytic method could enable investigators to explore changes within several networks to address a single question. In a recent study by Nomura *et al*[11] dissociation between two networks has been demonstrated in a group of chronic stroke patients with heterogeneous lesions. In their work, functional connectivity between and within two cognitive control networks (fronto-parietal and cingulo-opercular) was compared in patients with heterogeneous lesions affecting one network more than the other. They found decreased functional connectivity within the damaged, more than the undamaged, networks. Building on this line of research, our aims in the current study were to investigate whether this dissociation is generalizable, extending to all large-scale networks and to explore the feasibility of a global, whole-brain approach that can account for heterogeneous lesions across patients. Based on previously published empirical data[9,13,18–20] and computational models of stroke,[21,22] we hypothesized that lesions will result in larger alterations in functional connectivity over time in affected versus unaffected networks, demonstrating the generalizability of this phenomenon.

Here, we present a novel, network-based approach to study the longitudinal effects of heterogeneous lesions on functional networks. We apply whole-brain spatial concordance as a measure of change in the connectivity over time (as initially described in Lohmann *et al*[23]), and find that concordance preferentially decreases in affected networks across a heterogeneous lesion population. This finding reflects the general influence of localized lesions on distant functionally connected regions, and demonstrates the feasibility and potential of the suggested analytic approach for investigating resting-state fMRI data in a heterogeneous stroke population.

[1]Berlin School of Mind and Brain, The Mind-Brain Institute, Humboldt University, Berlin, Germany; [2]Max Planck Institute for Human Cognitive and Brain Sciences, Leipzig, Germany; [3]Institute of Psychology, Humboldt University, Berlin, Germany; [4]Center for Stroke Research, Charité University, Berlin, Germany and [5]Department of Neurology, Charité University, Berlin, Germany. Correspondence: S Ovadia-Caro, MSc, Berlin School of Mind and Brain, Humboldt-Universität zu Berlin, Luisenstraße 56, Haus 1, 10117 Berlin, Germany or Dr DS Margulies, Max Planck Institute for Human Cognitive and Brain Sciences, StephanStraße 1A, 04103 Leipzig, Germany.
E-mail: smadar.ovadia@gmail.com or margulies@cbs.mpg.de
Sources of support: The study was funded by Minerva Stiftung, DFG (Berlin School of Mind and Brain), Einstein Foundation Berlin (The Mind-Brain Institute), BMBF (Center for Stroke Research Berlin), and the German Competence Net Stroke.
Received 27 November 2012; revised 19 March 2013; accepted 26 April 2013; published online 29 May 2013

MATERIALS AND METHODS

Subjects

Thirty-one patients diagnosed with ischemic stroke were initially recruited for the study as part of the *1000+ Study* (registered in http://www.clinicaltrials.org, NCT00715533).[24] Inclusion criteria were first ever ischemic stroke within 24 hours, which was evident in imaging, and a minimum of three consecutive resting-state fMRI scans. Twelve patients were included in the current analysis due to exclusion criteria, including: antecedent lesions (5 excluded), brainstem or cerebellar infarcts, extensive white matter lesions defined as Wahlund score[25] $\geqslant 6$ (8 excluded), other brain abnormalities, revealed in anatomical scans (2 excluded), and less than three resting-state scans (4 patients), leaving 12 patients (age 64.25 ± 12.0 years, 8 males, 4 females) for the analysis. This study was approved by the ethics committee of the Charité-Universitätsmedizin, Berlin, Germany. Written informed consent was obtained from all patients.

Functional Imaging

Functional magnetic resonance imaging (fMRI) data were obtained during a 5.75-minutes (150 volumes) resting-state scan (i.e., spontaneous blood oxygenation level-dependent fluctuations) using a Siemens Tim Trio 3T scanner (Siemens Ag, Erlangen, Germany) at the Center for Stroke Research at the Charité University Hospital in Berlin. Patients were scanned at three consecutive time points after the stroke: day 1 post-stroke (1 day $\pm$ 0, mean $\pm$ std), day 7 post-stroke (8.25 ± 6.34 days), and day 90 post-stroke (90.12 ± 5.0 days). Day 1 was defined as the interval between 24 and 48 hours post-symptoms onset. The early acquisition was facilitated by the proximity of the scanner to the stroke unit. Two-dimensional functional images using blood oxygenation level-dependent contrast were obtained with an EPI sequence (TR = 2300 milliseconds, TE = 30 milliseconds, 30 slices, voxel size: $3.125\,\text{mm} \times 3.125\,\text{mm} \times 4\,\text{mm}$, flip angle 90°). T1-weighted anatomical images were acquired using a 3D MPRAGE sequence (TR = 1900 milliseconds, TE = 2.52 milliseconds, TI = 900 milliseconds, 192 slices, voxel size: $1\,\text{mm} \times 1\,\text{mm} \times 1\,\text{mm}$, flip angle 9°). Diffusion weighted images (DWI) and fluid attenuated inversion recovery (FLAIR) images acquired at day 1 post-stroke were used for lesion localization (in one patient, for which this data were not available for day 1 post-stroke, data from day 7 post-stroke were used). For further details on patients and scanning time points, see Supplementary Table S1.

fMRI Preprocessing

fMRI data were preprocessed using FSL (FMRIB Software Library, http://www.fmrib.ox.ac.uk) and AFNI (Analysis of Functional NeuroImages, http://afni.nimh.nih.gov/afni) software, based on the 1000 Functional Connectomes scripts (http://fcon_1000.projects.nitrc.org/). The first two images of each functional scan were discarded to avoid T1 saturation effects. Preprocessing of functional scans included: slice-time correction, 3D motion correction, spatial smoothing with a 6 mm full-width-at-half-maximum Gaussian kernel, band-pass filtering (0.009 to 0.1 Hz), removal of linear and quadratic trends and mean-based intensity normalization of all volumes by the same factor (10,000). Several sources of spurious variance were removed from the signal time-course of each voxel using the general linear model: global signal (average signal over the whole-brain mask), white matter signal, signal from the ventricles, and the six motion parameters. Mean motion displacement and rotation were computed for each patient and for each scan as described in Van Dijk *et al*.[26] Mean motion did not exceed a maximum of 2.5 mm in mean total displacement and 2.5 degrees in mean total rotation in any of the scans. For further details on patients' motion see Supplementary Table S2. We have made the scripts publically available: (https://github.com/NeuroanatomyAndConnectivity/checkMotion). To verify that global signal removal had not influenced the results, we repeated our analysis without this preprocessing step, and observed similar results (for further details, see Supplementary Material M1 and Supplementary Figure S1). Data were then normalized to the Montreal Neurologic Institute MNI152 template with $3\,\text{mm}^3$ resolution using FMRIB's Linear Image Registration Tool.[27,28] Each high-resolution structural image was registered to the MNI152 template by computing a 12 degrees-of-freedom linear affine transformation. Registration of each corresponding functional data to the high-resolution structural image was carried out using a linear transformation with 6 degrees-of-freedom. The structural-to-standard nonlinear transformation matrices were then applied to obtain a functional volume in MNI152 standard space.

Lesion Definition

Lesions were defined based on the FLAIR/DWI image acquired at day 1 post-stroke. Lesions were localized individually based on hyperintensity in the image, in addition to an independent radiologist report. Lesions were manually drawn on the image in native space using drawing tools available in FSLview. Registration of each FLAIR/DWI image to the high-resolution structural image was carried out using a linear transformation with 6 degrees-of-freedom. Each high-resolution structural image was registered to the MNI152 template by computing a 12 degree-of-freedom linear affine transformation. The structural-to-standard nonlinear transformation matrices were then applied, using the nearest neighbor interpolation (to avoid 'expansion' of the lesion area), to obtain a registered lesion mask in MNI152 standard space. These standard-space lesion masks were used for further computation of affected and unaffected networks based on a template set of networks. Normally, in the acute phase, DWI images are the most sensitive images for lesion delineation.[29] In our study, images were acquired 24 to 48 hours post-symptoms onset and in most cases, lesions were fully visible in the FLAIR images as well. DWI images were used for first inspection of hyperintensity localization. In the case of a complete match between the hyperintensity evident in the DWI image and the FLAIR image, the FLAIR was used for drawing the mask. In cases where lesions were not fully visible in the FLAIR image ($N = 2$), the mask was drawn on the DWI image. This was done since FLAIR images generally yielded better registration results to MNI152 standard-space.

Eight-Network Template

The eight-network template was taken from Beckmann *et al*[30] (available at: http://www.fmrib.ox.ac.uk/analysis/royalsoc8/). In the current study, the template was used for lesion mapping into affected and unaffected networks, as well as for functional connectivity analysis. Beckmann and colleagues computed a probabilistic independent component analysis[31] using resting-state fMRI data from 10 healthy controls. Probabilistic independent component analysis was used to characterize the spatiotemporal structure of the data. Their results demonstrate high spatial consistency between subjects as well as functional relevance of the revealed networks, which include areas of the visual cortex, sensory and motor systems. Our decision to use this template was based on the functional relevance of the independent components of the template as well as the high consistency across subjects.[30,32] Based on the publication by Beckmann *et al*,[30] the eight-network template can be functionally defined as follows: (a) medial visual cortical areas, (b) lateral visual cortical areas, (c) auditory system, (d) sensorimotor system, (e) visuospatial system (i.e., default-mode network[33]), (f) executive control, (g, h) right and left dorsal visual stream (respectively). For a detailed description of the anatomical structures in each of the networks, see Beckmann *et al*.[30]

To verify that results were not dependent on the choice of network templates, we repeated the analysis using a set of 20 networks from Smith *et al*.[32,34] This analysis yielded similar results to those found for the eight-network template (for further details see Supplementary Material M2, Supplementary Figure S2, and Supplementary Figure S3).

Defining Networks Affected by Lesions

To classify the eight-networks into affected/unaffected individually, an overlap between the binarized eight-networks thresholded template and the individual lesion masks was computed through multiplication. An overlap (minimum of one voxel) resulted in assigning that network as affected; no overlap resulted in assigning that network as unaffected. This yielded a vector of eight networks for each patient. In this vector, values of 0 reflect unaffected network by the individual lesion, and values of 1 reflect affected networks. For a description of lesion sites, see Supplementary Table S1.

Analysis of Functional Connectivity

Functional connectivity maps for each individual functional scan were computed using dual-regression.[35,36] Spatial maps based on a group-ICA of the eight-network template were first used as a set of general linear model regressors on the individual-level. This resulted in a time-course describing the similarity of each volume to the independent component template. These time-courses were then used as general linear model regressors in a second multiple regression analysis, resulting in individual-level spatial maps for each original component. For an example of representative individualized spatial maps see Supplementary Material M3 and Supplementary Figure S4.

To quantify the spatial similarity over time points, we computed the spatial concordance for each patient across time points, such that each component resulted in a single value representing spatial similarity of the network over time. Concordance was computed using concordance correlation coefficient[37] in MATLAB R2011a (The MathWorks Inc., Natick, MA, USA). Concordance values range from 1 (no difference) to -1 (maximal difference). Concordance was computed on thresholded spatial maps using a *z*-value $\geqslant 2.3$ (corresponds to a *p*-value of 0.01). We previously applied a similar approach on the voxel-wise level for exploratory analysis of longitudinal resting-state data from a patient following stroke.[23] The method was extended here to assess changes in concordance at the network level (rather than at the voxel level).

To exclude the lesion area from spatial concordance computation, the individual lesion masks were smoothed (using fslmaths –dilM) and excluded from concordance computation (by means of subtraction from the individual patients maps). This was done to verify that changes in the spatial pattern of functional connectivity maps could not be attributed to changes in the lesion area itself.

To test whether concordance values statistically differed in affected versus unaffected networks, we computed Δ-concordance for each patient: averaging the concordance values for the affected and unaffected networks separately, and then subtracting the averages: $((\mu_{\text{unaffected}}) - (\mu_{\text{affected}}))$. A one-sample *t*-test was used to test for significant difference between concordance in affected and unaffected networks. Figure 1 provides a schematic illustration of the different analysis steps.

Analysis of Clinical Data

In order to investigate the link between changes in functional connectivity as measured by concordance and behavioral change over time, we have used clinical data from day 1 and day 90, as measured by the National Institute of Health Stroke Scale (NIHSS). A measure of Δ-NIHSS was computed and correlated with Δ-concordance by means of Spearman's correlation coefficient. Δ-NIHSS was computed as the ranked absolute difference between the NIHSS obtained at day 1, and the NIHSS obtained at day 90. Higher values thus reflect larger clinical change over time.

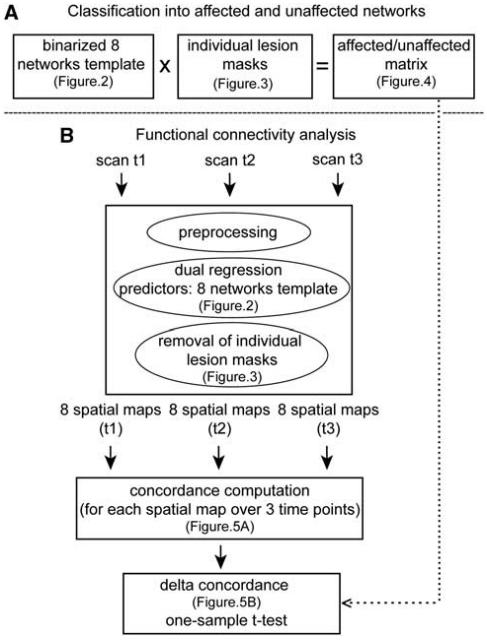

Figure 1. A flow chart of analysis steps. (**A**) Classification of individual lesions into affected and unaffected networks. The binarized eight-network template was multiplied by each individual lesion mask to classify lesions to affected/unaffected matrix. (**B**) Functional connectivity analysis. Each scan was preprocessed and dual-regression was performed. After removal of individual lesion masks, spatial concordance correlation coefficient was computed for each map over three time points. Δ-concordance $((\mu_{\text{unaffected}}) - (\mu_{\text{affected}}))$ was computed and a one-sample *t*-test was performed.

RESULTS

Lesion Mapping: The Network Approach

To map heterogeneous lesions across our sample of patients, we computed the overlap between the eight independent networks (Figure 2) and individual lesions (Figure 3). The overlap result, shown in Figure 4, represents the affected and unaffected networks for each patient's lesion. For example, the lesion for patient 1 was located in the left putamen and left insular cortex. According to the network approach applied here, the networks affected by the lesion were c, d, and f. For patient 2, the lesion was larger in size and was located in the left postcentral gyrus and the left parietal operculum extending into the supramarginal gyrus. The affected networks were networks b–h. For patient 3, the lesion was located in the left pallidum extending to some parts of the left amygdala. Only network c was affected.

The Impact of a Lesion on Functional Connectivity

To explore the influence of a lesion on functional connectivity over time, we computed a measure of spatial concordance of each network, for each patient, across all time points. Figure 5A depicts individual concordance values for each network of each patient. Networks with high concordance values reflect little change over time in the spatial pattern of the functional connectivity maps. After concordance computation, networks were assigned into affected and unaffected as described earlier (see Figure 4). Figure 5B displays the individual Δ-concordance values for each patient. Positive values reflect higher average concordance in unaffected networks, as compared with affected networks. Using a one-sample *t*-test, we found a significant difference ($P = 0.018$, one-sided), indicating that affected networks were significantly less concordant than unaffected networks. This reflects a stronger change in functional connectivity over time in affected networks. We accounted for potential contamination due to changes within the lesion area by excluding it from the analysis.

The Relationship Between Concordance and Clinical Change

To explore the clinical significance of our findings, a correlation between Δ-NIHSS and Δ-concordance was computed using Spearman's correlation coefficient. As alterations in functional connectivity after stroke have been previously shown to correlate with behavior,[9,13,18–20] we hypothesized a positive correlation between the two measures, such that the larger the difference between unaffected and affected networks, the larger the clinical change. As can be seen in Figure 6, a significant ($P = 0.05$, one-sided) positive correlation ($r = 0.5$) was found between the two measures, providing support for the clinical significance of our measure.

DISCUSSION

We found that the changes induced by lesions preferentially impact its functional networks. In order to address this question, we developed a novel analytic approach capable of assessing network-based changes in a heterogeneous stroke population. A generalizable analysis was necessary to investigate whether the effects of localized stroke are relevant to distributed, but interconnected areas, irrespective of the lesion location or implicated

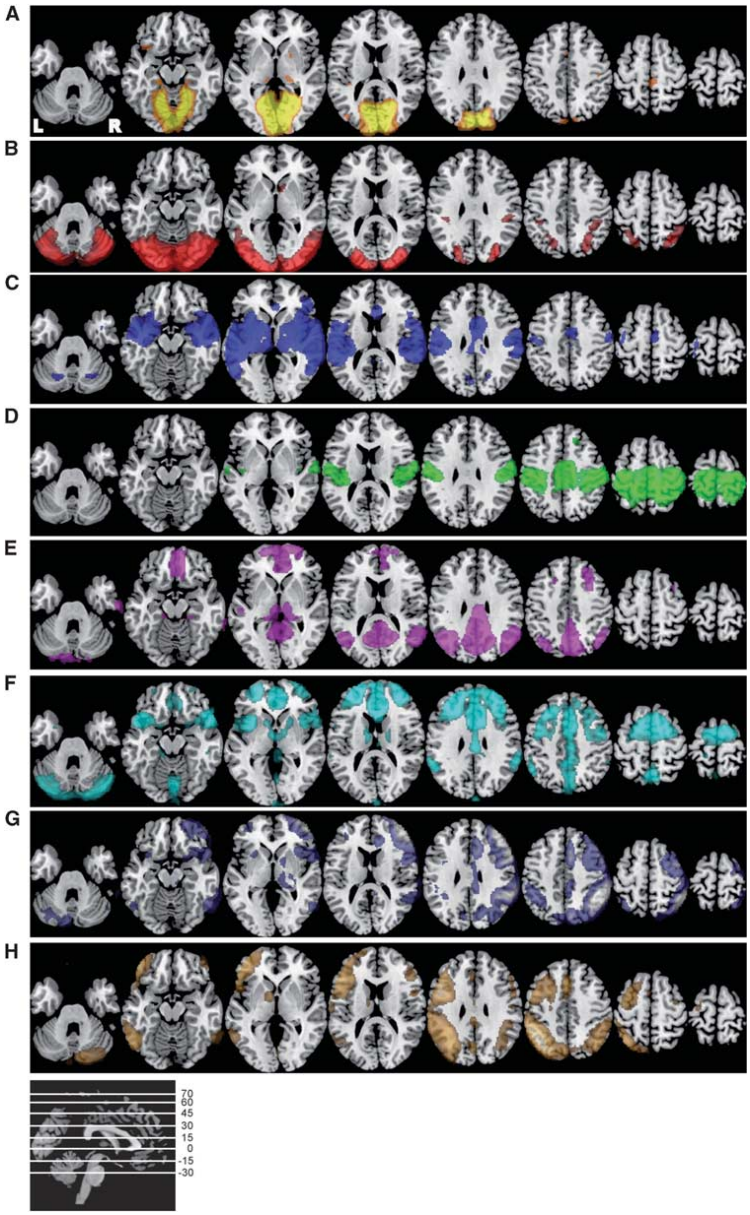

Figure 2. The eight-network template. Axial slices of the eight independent components based on probabilistic independent component analysis in healthy controls ($N = 10$) adopted from Beckmann *et al.*[30] This template was used as a basis for computation of overlap between lesions and networks as well as for dual-regression analysis. Images are shown in neurological convention, *z*-coordinates can be seen on sagittal view. Maps are thresholded based on histogram mixture modeling as described in Beckmann *et al.*

network. Beginning with data acquired 1 day post-stroke onset, we found that networks containing lesions decreased significantly in their concordance over time compared with the unaffected networks. In addition, a significant positive correlation was found between the clinical change over time and alteration in functional connectivity, as measured by Δ-concordance.

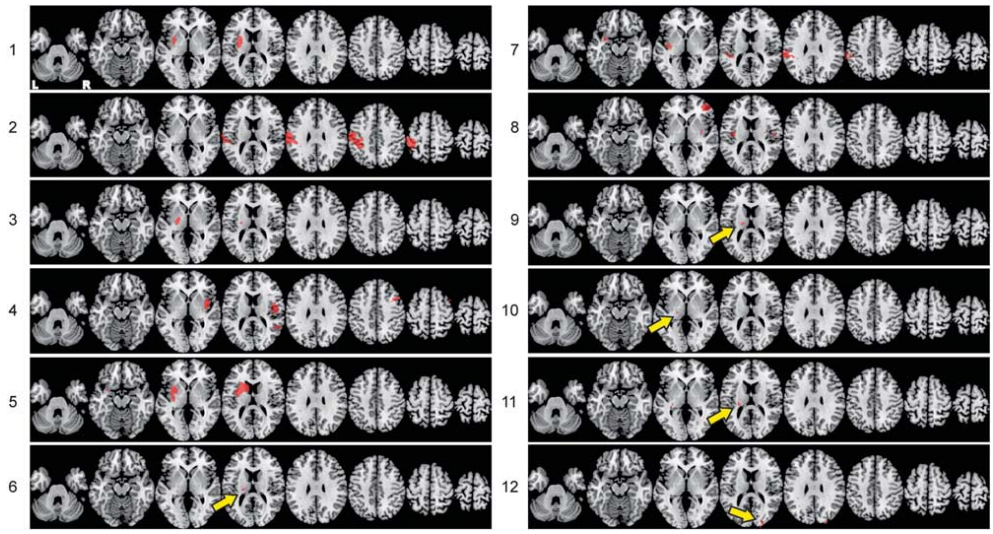

Figure 3. Individualized lesion masks. Axial slices of the individual lesions based on DWI/FLAIR images registered to standard MNI152 space. *z*-coordinates are identical to those presented in Figure 2. Red color depicts areas of hyper-intensity, i.e., the lesioned areas. Yellow arrows are pointing to small lesion areas to improve visibility.

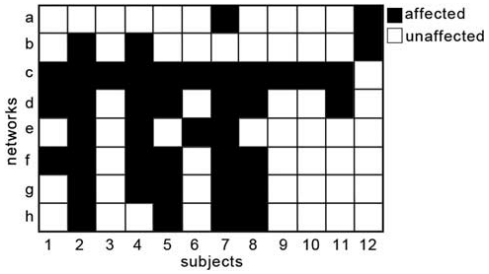

Figure 4. Affected and unaffected networks. Based on the overlap between the eight-network template and the individualized lesion masks, networks were assigned into affected (black) or unaffected (white) for each patient (x-axis). The y-axis depicts individual networks as presented in Figure 2.

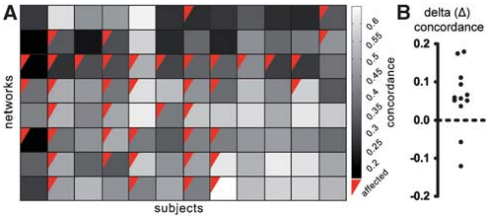

Figure 5. Spatial concordance in affected and unaffected networks. (**A**) Spatial concordance as computed over time for each patient (x-axis) and for each network (y-axis). High values reflect a small change in the spatial pattern. Red triangles depict affected networks. (**B**) Δ-concordance $((\mu_{\text{unaffected}}) - (\mu_{\text{affected}}))$ was computed for each patient demonstrating a significant positive distribution as tested by one-sample *t*-test.

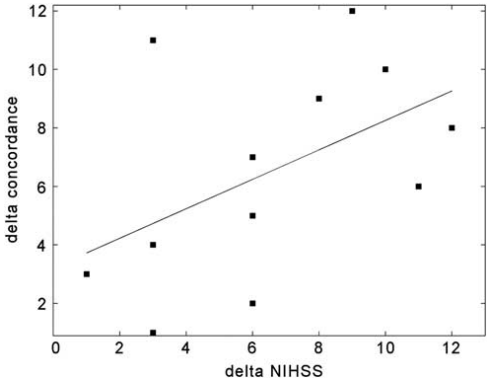

Figure 6. Relationship between Δ-concordance and clinical change. Positive correlation between changes in clinical scores over time as measured by Δ-NIHSS (x-axis) and changes in functional connectivity as measured by Δ-concordance (y-axis). Both axes depict the ranked values (as Spearman's correlation was applied to statistically test the relationship). Black line depicts the fitted regression line.

This correlation provides support for the clinical significance of our findings.

Our results are in line with previous work demonstrating that alterations in functional connectivity after stroke extend beyond the lesion area. For example, He *et al* [9] reported a breakdown of interhemispheric functional connectivity within the attention network in stroke patients with neglect symptoms. Symptom severity correlated with decreased connectivity, and recovery from symptoms correlated with the recovery of normal connectivity patterns. Similar results have been reported in the

sensorimotor[12,13,18,20,38] and language networks.[39] In addition to the well-established correlation between functional connectivity and behavioral symptoms, intervention/treatment has been shown to normalize functional connectivity patterns,[40–42] emphasizing the potential usefulness of functional connectivity as a surrogate measure of functional recovery and for the assessment of rehabilitation tools (for detailed reviews, see Carter *et al*[43] and Grefkes and Fink[44]).

Alterations of functional connectivity in heterogeneous lesions have been previously explored by Nomura *et al*.[11] Patients were examined using a single scan at least 5 months post-stroke or injury. The results demonstrated double dissociation of two cognitive control networks (frontoparietal and cingulo-opercular). Functional connectivity was decreased within the damaged networks more than within the undamaged network. While in Nomura *et al* the network approach was the basis for supporting independence between two networks, our results extend these findings to involve networks falling outside of the lesion area across the whole brain. In addition, Nomura *et al* addressed a chronic stroke population using a single scan, while we were interested in the functional change over time. While Nomura *et al* established the relation between lesions and functional connectivity-based networks, it was our aim to expand such findings to a generalized understanding of changes in network recovery over time.

Owing to the challenge of acquiring longitudinal data in a stroke population, only three studies have addressed the dynamics of functional connectivity during rest after stroke in humans,[13,18,19] all addressing recovery within the motor network. Park and colleagues[18] explored dynamics within the ipsilesional primary motor area and found decreased interhemispheric connectivity, which was most prominent 1-month post-stroke onset. Wang and colleagues[13] used graph theory to describe a gradual shift towards a random graph structure, suggesting a less-effective network state. They explored changes starting at 1-week post-stroke onset. Recently, Golestani and colleagues[19] have addressed the longitudinal recovery after stroke starting the acute phase (<24 hours). They demonstrated decreased interhemispheric connectivity within the motor network at the acute phase, which recovered 7 days post-stroke in recovered patients. Their work is the first longitudinal stroke study beginning in the acute phase. Their results demonstrate the importance of acute resting-state data to capture early changes in functional connectivity and behavioral outcome after stroke.

Experimental stroke research in rats has also demonstrated similar results to those reported in humans.[45–47] Van Meer et al[45] explored the longitudinal effect of unilateral stroke on the sensorimotor system in rats and found decreased interhemispheric and increased intrahemispheric synchronization. Over the course of recovery, reorganization and relative normalization of functional connectivity correlated with behavior.

The measure of concordance presented here reflects a change in functional connectivity and cannot explain directionality or the source of change within a network. One of the potential limitations of concordance is that certain networks are generally more stable over time than others.[23,48] For example, Lohmann *et al*, who proposed the use of voxel-wise concordance with functional connectivity data, found brain areas that were more concordant than others in healthy controls.[23] However, in our analysis, concordance is computed for each subject individually, producing one single value for each network over time. This decreases the potential influence of differences in levels of concordance between subjects. In addition, the fact that our sample is heterogeneous, with lesions distributed across different networks, further reduces the potential confound for within-subject comparison.

The relationship between alterations in functional connectivity after stroke and behavioral outcome/performance has been previously reported for specific networks.[9,13,18–20] In our study, although the clinical significance of our findings should be further explored by examining the link between specific clinical outcome in the different networks (with their respective domains) and concordance, our results support the theoretical framework concerning the influence of a lesion on a complex network of interconnected brain regions. This view is supported as well by the well-known phenomenon of diaschisis[49] in which, after stroke, regions far from the lesion site can show altered metabolic and neural activity. Changes in functional connectivity in general and our suggested approach in particular can be used to shed light on this phenomenon.

The study of longitudinal effects of stroke on functional networks in heterogeneous stroke populations can contribute to our understanding of intrinsic networks organization in the human brain. By providing insight into the recovery process after stroke, future research into large-scale networks may prove valuable for rehabilitation and prognosis.

DISCLOSURE/CONFLICT OF INTEREST

The authors declare no conflict of interest.

ACKNOWLEDGEMENTS

We thank Dr Karsten Müller, Dr Guido Hesselmann, Dr Fatma Imamoglu, and Dr Gabriele Lohmann for their help and advice in various stages of this project.

REFERENCES

1 Sporns O. From simple graphs to the connectome: networks in neuroimaging. *Neuroimage* 2011; **62**: 881–886.
2 Sporns O, Chialvo DR, Kaiser M, Hilgetag CC. Organization, development and function of complex brain networks. *Trends Cogn Sci* 2004; **8**: 418–425.
3 Bullmore E, Sporns O. The economy of brain network organization. *Nat Rev Neurosci* 2012; **13**: 336–349.
4 Bullmore E, Sporns O. Complex brain networks: graph theoretical analysis of structural and functional systems. *Nat Rev Neurosci* 2009; **10**: 186–198.
5 Ovadia-Caro S, Nir Y, Soddu A, Ramot M, Hesselmann G, Vanhaudenhuyse A *et al.* Reduction in inter-hemispheric connectivity in disorders of consiousness. *PLoS ONE* 2012; **7**: e37238.
6 Boly M, Massimini M, Garrido MI, Gosseries O, Noirhomme Q, Laureys S *et al.* Brain connectivity in disorders of consciousness. *Brain Connect* 2012; **2**: 1–10.
7 Buckner RL, Sepulcre J, Talukdar T, Krienen FM, Liu H, Hedden T *et al.* Cortical hubs revealed by intrinsic functional connectivity: mapping, assessment of stability, and relation to Alzheimer's disease. *J Neurosci* 2009; **29**: 1860–1873.
8 Greicius M. Resting-state functional connectivity in neuropsychiatric disorders. *Curr Opin Neurol* 2008; **21**: 424–430.
9 He BJ, Snyder AZ, Vincent JL, Epstein A, Shulman GL, Corbetta M. Breakdown of functional connectivity in frontoparietal networks underlies behavioral deficits in spatial neglect. *Neuron* 2007; **53**: 905–918.
10 He BJ, Shulman GL, Snyder AZ, Corbetta M. The role of impaired neuronal communication in neurological disorders. *Curr Opin Neurol* 2007; **20**: 655–660.
11 Nomura EM, Gratton C, Visser RM, Kayser A, Perez F, D'Esposito M. Double dissociation of two cognitive control networks in patients with focal brain lesions. *Proc Natl Acad Sci USA* 2010; **107**: 12017–12022.
12 Grefkes C, Nowak DA, Eickhoff SB, Dafotakis M, Kust J, Karbe H *et al.* Cortical connectivity after subcortical stroke assessed with functional magnetic resonance imaging. *Ann Neurol* 2008; **63**: 236–246.
13 Wang L, Yu C, Chen H, Qin W, He Y, Fan F *et al.* Dynamic functional reorganization of the motor execution network after stroke. *Brain* 2010; **133**(Pt 4): 1224–1238.
14 Rossini PM, Calautti C, Pauri F, Baron JC. Post-stroke plastic reorganisation in the adult brain. *Lancet Neurol* 2003; **2**: 493–502.
15 Hillis AE, Barker PB, Beauchamp NJ, Gordon B, Wityk RJMR. perfusion imaging reveals regions of hypoperfusion associated with aphasia and neglect. *Neurology* 2000; **55**: 782–788.
16 Hillis AE, Gold L, Kannan V, Cloutman L, Kleinman JT, Newhart M *et al.* Site of the ischemic penumbra as a predictor of potential for recovery of functions. *Neurology* 2008; **71**: 184–189.
17 Feeney DM, Baron JC. Diaschisis. *Stroke* 1986; **17**: 817–830.

18 Park CH, Chang WH, Ohn SH, Kim ST, Bang OY, Pascual-Leone A *et al.* Longitudinal changes of resting-state functional connectivity during motor recovery after stroke. *Stroke* 2011; **42**: 1357–1362.
19 Golestani AM, Tymchuk S, Demchuk A, Goodyear BG. Longitudinal evaluation of resting-state FMRI after acute stroke with hemiparesis. *Neurorehabil Neural Repair* 2013; **27**: 153–163.
20 Carter AR, Astafiev SV, Lang CE, Connor LT, Rengachary J, Strube MJ *et al.* Resting interhemispheric functional magnetic resonance imaging connectivity predicts performance after stroke. *Ann Neurol* 2010; **67**: 365–375.
21 Alstott J, Breakspear M, Hagmann P, Cammoun L, Sporns O. Modeling the impact of lesions in the human brain. *PLoS Comput Biol* 2009; **5**: e1000408.
22 Honey CJ, Sporns O. Dynamical consequences of lesions in cortical networks. *Hum Brain Mapp* 2008; **29**: 802–809.
23 Lohmann G, Ovadia-Caro S, Jungehulsing GJ, Margulies DS, Villringer A, Turner R. Connectivity concordance mapping: a new tool for model-free analysis of FMRI data of the human brain. *Front Syst Neurosci* 2012; **6**: 13.
24 Hotter B, Pittl S, Ebinger M, Oepen G, Jegzentis K, Kudo K *et al.* Prospective study on the mismatch concept in acute stroke patients within the first 24 h after symptom onset—1000Plus study. *BMC Neurol* 2009; **9**: 60.
25 Wahlund LO, Barkhof F, Fazekas F, Bronge L, Augustin M, Sjogren M *et al.* A new rating scale for age-related white matter changes applicable to MRI and CT. *Stroke* 2001; **32**: 1318–1322.
26 Van Dijk KR, Sabuncu MR, Buckner RL. The influence of head motion on intrinsic functional connectivity MRI. *Neuroimage* 2012; **59**: 431–438.
27 Jenkinson M, Bannister P, Brady M, Smith S. Improved optimization for the robust and accurate linear registration and motion correction of brain images. *Neuroimage* 2002; **17**: 825–841.
28 Jenkinson M, Smith S. A global optimisation method for robust affine registration of brain images. *Med Image Anal* 2001; **5**: 143–156.
29 Luby M, Bykowski JL, Schellinger PD, Merino JG, Warach S. Intra- and interrater reliability of ischemic lesion volume measurements on diffusion-weighted, mean transit time and fluid-attenuated inversion recovery MRI. *Stroke* 2006; **37**: 2951–2956.
30 Beckmann CF, DeLuca M, Devlin JT, Smith SM. Investigations into resting-state connectivity using independent component analysis. *Philos Trans R Soc Lond B Biol Sci* 2005; **360**: 1001–1013.
31 Beckmann CF, Smith SM. Probabilistic independent component analysis for functional magnetic resonance imaging. *IEEE Trans Med Imaging* 2004; **23**: 137–152.
32 Laird AR, Fox PM, Eickhoff SB, Turner JA, Ray KL, McKay DR *et al.* Behavioral interpretations of intrinsic connectivity networks. *J Cogn Neurosci* 2011; **23**: 4022–4037.
33 Raichle ME, MacLeod AM, Snyder AZ, Powers WJ, Gusnard DA, Shulman GL. A default mode of brain function. *Proc Natl Acad Sci USA* 2001; **98**: 676–682.
34 Smith SM, Fox PT, Miller KL, Glahn DC, Fox PM, Mackay CE *et al.* Correspondence of the brain's functional architecture during activation and rest. *Proc Natl Acad Sci USA* 2009; **106**: 13040–13045.
35 Beckmann CF, Mackay CE, Filippini N, Smith S. Group comparison of resting-state FMRI data using multi-subject ICA and dual regression. *Fifteenth Annual Meeting of the Organization for Human Brain Mapping* 2009.
36 Filippini N, MacIntosh BJ, Hough MG, Goodwin GM, Frisoni GB, Smith SM *et al.* Distinct patterns of brain activity in young carriers of the APOE-epsilon4 allele. *Proc Natl Acad Sci USA* 2009; **106**: 7209–7214.
37 Lin LI. A concordance correlation coefficient to evaluate reproducibility. *Biometrics* 1989; **45**: 255–268.
38 Carter AR, Patel KR, Astafiev SV, Snyder AZ, Rengachary J, Strube MJ *et al.* Upstream dysfunction of somatomotor functional connectivity after corticospinal damage in stroke. *Neurorehabil Neural Repair* 2012; **26**: 7–19.
39 Warren JE, Crinion JT, Lambon Ralph MA, Wise RJ. Anterior temporal lobe connectivity correlates with functional outcome after aphasic stroke. *Brain* 2009; **132**(Pt 12): 3428–3442.
40 Nowak DA, Grefkes C, Ameli M, Fink GR. Interhemispheric competition after stroke: brain stimulation to enhance recovery of function of the affected hand. *Neurorehabil Neural Repair* 2009; **23**: 641–656.
41 Hummel FC, Cohen LG. Non-invasive brain stimulation: a new strategy to improve neurorehabilitation after stroke? *Lancet Neurol* 2006; **5**: 708–712.
42 Grefkes C, Nowak DA, Wang LE, Dafotakis M, Eickhoff SB, Fink GR. Modulating cortical connectivity in stroke patients by rTMS assessed with fMRI and dynamic causal modeling. *Neuroimage* 2010; **50**: 233–242.
43 Carter AR, Shulman GL, Corbetta M. Why use a connectivity-based approach to study stroke and recovery of function? *Neuroimage* 2012; **62**: 2271–2280.
44 Grefkes C, Fink GR. Reorganization of cerebral networks after stroke: new insights from neuroimaging with connectivity approaches. *Brain* 2011; **134** (Pt 5): 1264–1276.
45 van Meer MP, van der Marel K, Wang K, Otte WM, El Bouazati S, Roeling TA *et al.* Recovery of sensorimotor function after experimental stroke correlates with restoration of resting-state interhemispheric functional connectivity. *J Neurosci* 2010; **30**: 3964–3972.
46 van Meer MP, van der Marel K, Otte WM, Berkelbach van der Sprenkel JW, Dijkhuizen RM. Correspondence between altered functional and structural connectivity in the contralesional sensorimotor cortex after unilateral stroke in rats: a combined resting-state functional MRI and manganese-enhanced MRI study. *J Cereb Blood Flow Metab* 2010; **30**: 1707–1711.
47 van Meer MP, Otte WM, van der Marel K, Nijboer CH, Kavelaars A, van der Sprenkel JW *et al.* Extent of bilateral neuronal network reorganization and functional recovery in relation to stroke severity. *J Neurosci* 2012; **32**: 4495–4507.
48 Shehzad Z, Kelly AM, Reiss PT, Gee DG, Gotimer K, Uddin LQ *et al.* The resting brain: unconstrained yet reliable. *Cereb Cortex.* 2009; **19**: 2209–2229.
49 Finger S, Koehler PJ, Jagella C. The Monakow concept of diaschisis: origins and perspectives. *Arch Neurol* 2004; **61**: 283–288.

Supplementary Information accompanies the paper on the Journal of Cerebral Blood Flow & Metabolism website (http://www.nature.com/jcbfm)

Topical Review

Section Editors: Wolf-Dieter Heiss, MD, and Chelsea S. Kidwell, MD

The Value of Resting-State Functional Magnetic Resonance Imaging in Stroke

Smadar Ovadia-Caro, MSc; Daniel S. Margulies, PhD; Arno Villringer, MD

In the acute phase of stroke, the use of imaging techniques aims to provide pathophysiological information concerning vascular patency, areas of hypoperfusion, and metabolic and structural damage. Based on such information, therapeutic decisions such as the administration of reperfusion medications are made. After the acute phase, brain plasticity and reorganization are the main mechanisms underlying functional recovery, and improvement is determined by functional adaptations of distributed brain networks mediated by connectivity.[1] Accordingly, new therapeutic approaches, such as noninvasive brain stimulation, target the modulation of connectivity and network function.[2,3] At this stage, imaging-based biomarkers should reflect the status of cerebral networks. As the relevance of the network view of stroke becomes increasingly evident,[4] so does the usefulness of imaging techniques in the assessment of cerebral network function in clinical populations. Most notably is the use of resting-state functional MRI (rs-fMRI).

rs-fMRI is a task-independent functional neuroimaging approach based on intrinsic low-frequency fluctuations (typically <0.1 Hz) in the blood oxygenation level–dependent (BOLD) signal. This signal can be used to compute the temporal correlations between spatially remote areas, termed: functional connectivity. In the healthy brain, functional connectivity is increased between areas that are part of the same functional network even in the absence of task. The resulting spatial patterns closely resemble the activation patterns identified during specific tasks,[5] and these networks are referred to as resting-state networks.[6] Thus, rs-fMRI provides an approach for detailed investigation of functional networks, as well as a more general method for assessing changes in intrinsic neuronal activity. Unlike task-based methods, measures of intrinsic functional connectivity allow for flexible post hoc analyses that probe multiple functional networks. Additionally, the minimal demands on the patient during the scanning session make the technique an optimal choice for clinical settings.

rs-fMRI may offer the prospect of providing therapeutically useful information on both the focal vascular lesion and the connectivity-based reorganization and subsequent functional recovery. Here we provide an overview of recent applications of rs-fMRI to stroke diagnostics and prognostics and discuss future perspectives and considerations. We begin with methods used to characterize local alterations in acute stroke and proceed to describe studies of specific and general connectivity changes at various phases of the recovery process. For a detailed description of the studies reviewed here, see Table I in the online-only Data Supplement.

Local Intrinsic BOLD Activity as a Measure of Hypoperfusion

Correlation analyses based on the BOLD signal are thought to reflect neuronal synchronization.[7] However, the BOLD signal additionally contains information concerning local blood flow and oxygen consumption[8] and is, therefore, potentially useful for assessing pathophysiological events within the stroke lesion itself. Current stroke MRI approaches use MR angiography, fluid attenuated inversion recovery, as well as diffusion and perfusion imaging to identify the severely damaged infarct core and, most importantly, the potentially salvable tissue on appropriate reperfusion therapy. The necessity of susceptibility contrast agent application in perfusion imaging[9] is a major disadvantage because it can cause severe side effects (eg, nephrogenic systemic fibrosis). In addition, the use of a contrast agent prohibits the acquisition of repeated scans during the same session, which can be necessary in a clinical setting because of data loss (eg, from excessive motion). Alternative noninvasive approaches such as arterial spin labeling (ASL) have been suggested to replace contrast-based perfusion imaging.[10,11] ASL has the advantage of quantitatively assessing perfusion with no need of contrast agent. However, so far, it has not been widely used in clinical setting possibly because of low signal-to-noise ratio in areas with long transit times.[11] In a recent study, ASL failed to detect 7 of 39

Received February 10, 2014; final revision received June 3, 2014; accepted June 9, 2014.

From the Mind-Brain Institute at the Berlin School of Mind and Brain, Charité and Humboldt University, Berlin, Germany (S.O.-C., D.S.M., A.V.); Department of Neurology, Max Planck Institute for Human Cognitive and Brain Sciences, Leipzig, Germany (S.O.-C., D.S.M., A.V.); Institute of Psychology, Humboldt University, Berlin, Germany (S.O.-C.); Center for Stroke Research, Charité Universitätsmedizin Berlin, Berlin, Germany (A.V.); and Department of Cognitive Neurology, University Hospital, Leipzig, Germany (A.V.).

The online-only Data Supplement is available with this article at http://stroke.ahajournals.org/lookup/suppl/doi:10.1161/STROKEAHA.114.003689/-/DC1.

Correspondence to Smadar Ovadia-Caro, MSc, Department of Neurology, Max-Planck Institute for Human Cognitive and Brain Sciences, Stephanstrasse 1a, 04103, Leipzig, Germany. E-mail smadar.ovadia@gmail.com

(*Stroke*. 2014;45:2818-2824.)

perfusion lesions.[12] In contrast, recent developments in ASL may improve its applicability.[10]

rs-fMRI, which also does not require the use of a contrast agent, has recently been used to identify the perfusion deficit.[13] Using time shift analysis, a high spatial correspondence has been found on the individual level with the area of hypoperfusion as defined by perfusion imaging (see Figure 1).[13] Time shift analysis was defined as the temporal shift necessary for maximum correlation with an average representative time series (ie, the global mean). These findings have been replicated in both acute patients after stroke and patients with chronic stenoocclusive vessel disease.[14] Given these results of time delay from the global mean, rs-fMRI could provide comparable results to those of conventional perfusion MRI without the need for contrast agents and may be of clinical value for diagnostic decisions, even in the acute phase after stroke. Although promising, this recent discovery should be further validated in larger cohorts, and issues such as motion artifacts and correlation with different perfusion parameters (ie, mean transient time and time to peak) should be further explored. In addition, scanning time used to obtain rs-fMRI–based results was longer in the 2 studies as compared with contrast-based perfusion scan. However, one of the studies demonstrated that reducing scanning time to 184 seconds still yielded similar results to those obtained using a full-length scan.[13] It is yet to be determined whether modification of scanning parameters (eg, faster repetition time afforded by newly developed multiband pulse sequences)[15] may be used to reduce scanning time without compromising the quality of the results.

Alterations in rs-fMRI Connectivity After Stroke

Evidence from animal studies suggests that processes such as axonal sprouting after ischemic lesions are induced by intrinsic patterns of synchronous low-frequency neuronal activity in areas connected to the infarct core.[16] This physiological role of intrinsic synchronous activity in areas capable of compensating for lost function, such as interhemispheric homologues, may underlie poststroke changes in functional connectivity. The impact of stroke on intrinsic BOLD activity has been widely characterized by describing such alterations in functional connectivity. The general effect reported thus far is a decrease in functional connectivity in areas that are structurally intact yet are connected to the lesion area. This phenomenon has been widely demonstrated in single networks, usually using a relatively small number of regions of interest (ROIs). In addition, perhaps the most promising finding is that changes in functional connectivity after stroke have been shown to correspond with the degree of behavioral deficit, emphasizing the prognostic value of rs-fMRI in stroke patients.

The advancement in our understanding of stroke as a network disorder dependent on global whole-brain communication and internetwork interaction, along with the development of methods in the wider field of functional connectivity, has created a shift in the methodological approaches applied to the study of stroke. As we will discuss in the following sections, early studies predominantly addressed alterations in specific networks, whereas more recent studies describe global graph-based changes.

Network-Specific Effects of Stroke

The sensorimotor network has been the most widely studied thus far, with a focus on interhemispheric functional connectivity. Interhemispheric connectivity between homologous regions is one of the prominent characteristics of resting-state connectivity patterns in healthy population and provides a stable and robust measure for the integrity of communication between the 2 hemispheres.[17] Alterations in connectivity between the arm subregions of the sensorimotor network have been found to correlate with the upper extremity motor impairment in patients with hemiparesis.[18] A decrease in interhemispheric functional connectivity was additionally reported for patients with corticospinal tract damage, further supporting the fact that the reduction in functional connectivity after stroke cannot be solely explained by structural damage and reflects distant effects of the lesion in areas that remain intact.[19]

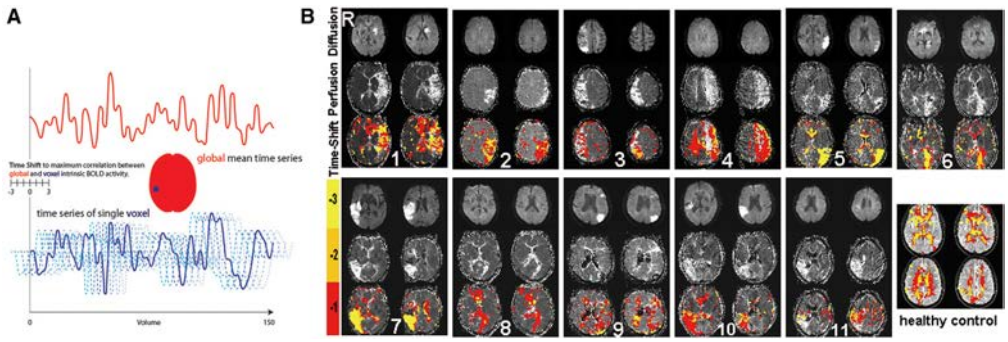

Figure 1. The resting-state functional MRI blood oxygenation level–dependent (BOLD) signal provides information similar to perfusion imaging. **A**, Time shift analysis. The time delay between the average whole-brain signal and each voxel was computed using time-lagged correlation. **B**, Areas of delayed BOLD from the global mean correspond to perfusion deficits, whereas diffusion depicts only the infarct core. Red-yellow scale colors reflect the delay in repetition time (TR). Adapted from Lv et al[13] with permission of the publisher. Copyright © 2013, John Wiley & Sons. Authorization for this adaptation has been obtained both from the owner of the copyright in the original work and from the owner of copyright in the translation or adaptation.

The importance of interhemispheric connectivity has been further demonstrated in longitudinal studies. A decrease in interhemispheric connectivity in the motor cortex has been reported for patients scanned 4× during a 6-month period poststroke. The decrease in interhemispheric connectivity was accompanied by an increase in connectivity between the motor cortex and ipsilesional frontal and parietal cortex.[20] The reduction in interhemispheric functional connectivity has been reported even in the early stages after stroke in patients with motor deficits. Interestingly, connectivity between hemispheres recovered 7 days poststroke only in patients with recovered motor function, although the reduction in connectivity with subcortical regions remained after 90 days.[21] Animal studies have found similar results to those reported in humans. van Meer et al[22] explored the longitudinal changes in functional connectivity in rats after unilateral experimental stroke. A reduction of interhemispheric connectivity was found soon after stroke and was correlated with the sensorimotor deficit. Recovery of interhemispheric connectivity was associated with behavioral improvement (see Figure 2). The alterations in functional connectivity were later demonstrated to result from corresponding alterations in structural connectivity as measured by tracer uptake (manganese-enhanced MRI). A decrease in interhemispheric functional connectivity was associated with a decrease in transcallosal tracer transfer, whereas an increase in intrahemispheric functional connectivity was associated with a local increase in the tracer uptake. These results provide further support for a structural connectivity mechanism underlying changes in functional connectivity.[23]

The interhemispheric imbalance reported in the sensorimotor network is in accordance with findings from task-based fMRI and is the basis for the usage of noninvasive transcranial magnetic stimulation techniques for the treatment of patients with stroke.[2] However, it is yet to be determined how such stimulation affects the resting-state functional connectivity in patients with stroke, because most studies have made use of task-based connectivity techniques.

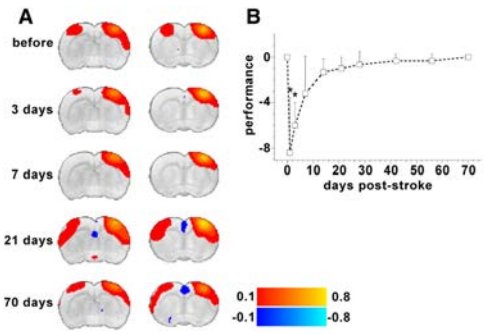

Figure 2. Correlation is decreased after stroke in areas that are structurally intact. Functional recovery correlates with the restoration of functional connectivity. **A**, Interhemispheric functional connectivity is reduced in structurally intact areas immediately after stroke and gradually recovers. Region of interest is located at the right forelimb region of the primary somatosensory cortex. **B**, Functional recovery as measured by sensorimotor performance is associated with recovery of functional connectivity. **P*<0.05 vs pre. Adapted from van Meer et al.[22] Copyright © 2010, The Authors (http://creativecommons.org/licenses/by-nc-sa/3.0/). Authorization for this adaptation has been obtained both from the owner of the copyright in the original work and from the owner of copyright in the translation or adaptation.

Similar results to those found in the sensorimotor network have been reported for the attention network. Damage to the attention network and the corresponding symptoms of spatial neglect have been associated with decreased interhemispheric connectivity in structurally intact areas that are part of the attention network.[18,24] Importantly, functional connectivity correlates with the severity of symptoms.[24] rs-fMRI has also been used to demonstrate the effect of intraparietal sulcus lesions on functional connectivity in the attention network. Depending on the location of the lesion within the intraparietal sulcus, functional connectivity was impaired, emphasizing the importance of the intraparietal sulcus in spatial attention, in addition to the well-established roles of the inferior parietal lobule and temporoparietal junction.[25]

Another network that has been explored after stroke is the default-mode network. The default-mode network is a network of regions including the posterior cingulate and precuneus, the temporoparietal junction, and the medial prefrontal cortex. It has been widely implicated in various neurological and mental disorders and has been linked to tasks such as autobiographical memory retrieval and theory of mind functions.[26] After stroke, alterations in default-mode network functional connectivity have been associated with poststroke depression[27] and episodic memory dysfunction.[28]

Generalizing the Network Impact of Stroke

Our understanding of the complexity of symptoms after stroke, which usually involves >1 network, and the importance of whole-brain connectivity has led to a gradual shift in the methods of analysis used in this clinical population. A shift from single-network assessment to a multinetwork and eventually whole-brain level is currently underway. The assessment of multiple domains and the interaction between them is necessary for the development of meaningful biomarkers to assess recovery and potentially prognosis. Nomura et al[29] were the first to provide an approach that could be applied to populations with heterogeneous lesions affecting >1 network. The aim of this work was to determine whether the frontoparietal and cinguloopercular networks are dissociated cognitive control networks and to test their independence. rs-fMRI data were collected ≥5 months poststroke, and functional connectivity was assessed across predefined ROIs within and between network nodes. The percentage of network damage was found to negatively correlate with functional connectivity within the affected network and not within the unaffected network.

We have extended the findings of Nomura et al[29] in a longitudinal study starting at the acute phase after stroke. We aimed to explore whether heterogeneous lesions to 8 a priori–defined spatial networks covering most of the cortical surface demonstrate stronger alterations in functional connectivity during the course of recovery as compared with unaffected networks at the individual level. Twelve patients with ischemic stroke were studied using rs-fMRI acquired 1, 7, and 90

days poststroke. Dual regression[30] was used to create functional connectivity maps for each of the predefined networks. We applied whole-brain spatial concordance to measure the changes in connectivity over time.[31] Our findings indicate a preferential decrease in concordance in networks affected by the lesion, as compared with unaffected networks. This finding reflects a more robust change in the functional connectivity spatial maps of the affected networks during the course of recovery. The change in connectivity was correlated with clinical changes as assessed by the National Institutes of Health Stroke Scale. Our results provide additional support for the generalization of diaschisis-like effects to patients with multiple network damage. In addition, we demonstrated the feasibility of our approach for the study of heterogeneous lesions.[32] Figure 3 is representing a schematic illustration of network disruption after stroke based on these empirical findings. A multinetwork assessment of changes in functional connectivity may have the potential of better reflecting the complex clinical symptoms after stroke.

Changes in Network Topology in Single Networks

Recently, methods from the mathematical field of graph theory have been applied to rs-fMRI data and structural data.[33] Although functional connectivity between predefined ROIs (seed-based ROI) has been proven valuable in investigating synchronization between specific regions, it does not provide us with information concerning the integrative ability of different regions, or nodes, within the network. Measures of graph theory can contribute to our understanding of topological organization of single and even multiple networks. Edges are determined based on connection described by correlation matrices. Various measures representing network structure and effectiveness can be computed, among them centrality, path length, clustering coefficient, and modularity.[33] Initially, measures of graph theory were used to study a single network after stroke, namely the motor network. In a 1-year longitudinal follow-up on patients having had subcortical stroke, a gradual shift in the motor network topology to a random mode has been reported, suggesting less efficient communication within the network. These changes were accompanied by a gradual increase in interhemispheric functional connectivity.[34] Interestingly, contradicting results were reported in a similar study conducted in rats. During the course of recovery, a gradual re-establishment of network properties was accompanied by normalization of interhemispheric functional connectivity.[35] These conflicting results may be explained by differences in scanning time, model type (ie, animal versus human), and bases for the graph reconstruction. In the study by Wang et al,[34] the network was built based on functional connectivity between ROIs, whereas in the study by van Meer et al,[35] the graph was computed for single voxels.

With the continuing advancement in computational capabilities, network properties can be examined using a larger number of regions, thereby creating more realistic graphs that better reflect the global properties of communication in the brain.[36]

Lesion Topology

Modeling studies on the impact of lesions on functional connectivity have provided similar results to those found in empirical data. The effect of simulated lesions extends beyond the immediate lesion environment to structurally intact areas. In addition, modeling studies suggest that the topological properties of the lesion itself have a meaningful effect on the

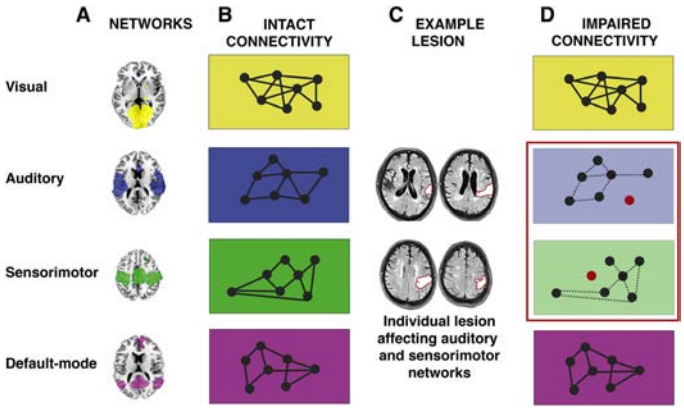

Figure 3. Schematic illustration of network disruption after stroke. **A**, Functional networks are correlated even in the absence of task. Here, an example of 4 functional networks based on resting-state functional MRI functional connectivity in healthy controls. **B**, Intact connectivity structure in healthy controls is reflected in the high correlation (solid black lines) between functionally relevant nodes of a specific network. **C**, Anatomic location of an individual lesion in a patient with a recent ischemic stroke (white areas outlined in red). The lesion affects the auditory and the sensorimotor networks, sparing the visual and default-mode networks. **D**, After stroke, structural damage to specific nodes (red circles) in the network leads to global disruption in connectivity in the affected networks (red rectangle), even in structurally intact regions. Connectivity is interrupted from the lesion area and altered among the structurally intact nodes of the network (black dotted lines). The disruption of connectivity after a local stroke is network-specific and largely spares the unaffected functional networks. A multinetwork assessment of changes in functional connectivity has better potential for reflecting complex clinical symptoms, which often involve >1 functional network.

amplitude of alterations in functional connectivity. More central connected regions (ie, hub regions) have a larger effect on functional connectivity after stroke.[37,38]

Hub regions can be defined by their role within the graph. Connector hubs connect different modules (eg, visual and motor network), whereas provincial hubs connect nodes within a single module (eg, within the visual network).[33] Gratton et al[39] empirically tested the influence of lesion topology on network integrity in patients after stroke using rs-fMRI. Whole-brain modularity was used as a measure of network integrity and was computed for both the affected and the unaffected hemispheres. Modularity was defined as a comparison between the number of connections within a module and the number of connections between different modules. In patients with stroke, a widespread decrease in modularity, even in the unaffected hemisphere, was found. The decrease in modularity was found to correlate with the increase in damage to connector hubs (high connector damage) and not to provincial hubs (low connector damage; Figure 4). The association between connector hub damage and modularity could not be explained by the lesion size alone. This study demonstrates the importance of connector hubs to the integrity of network structure; however, the link between hub damage and behavioral outcome after stroke is yet to be explored.

Conclusions, Considerations, and Future Perspectives

In summary, rs-fMRI has been successfully applied in patients with acute and chronic stroke. In acute stroke, time shift analysis based on rs-fMRI could potentially replace classic perfusion measurements without the need for contrast agent application. We are currently evaluating this approach in a larger clinical study. Using connectivity analysis based on rs-fMRI, focal infarcts have been shown to influence connectivity within the affected networks as well as disrupt whole-brain topology. These changes have been shown to correlate with behavioral measures as well as behavioral outcome.

Certain limitations should be taken into account when using rs-fMRI in patients with stroke. Functional connectivity is highly susceptible to motion-related artifacts, and because patients tend to move more than controls, there is a need for either real-time motion correction or improved post hoc removal of motion artifacts.[40] In addition, given that white matter lesions affect brain connectivity[41,42] and are reported to relate to behavioral deficits after stroke,[43] the variance explained by this factor needs to be accounted for when conducting functional connectivity analyses.

An additional challenge in using rs-fMRI is the interpretability of changes in the BOLD signal given the state of vascular pathology in patients with stroke. The BOLD signal mainly reflects changes in the concentration of deoxyhemoglobin and, as such, is an indirect measure of neuronal activity.[8,44] Comparing stroke patients' data to those obtained from healthy controls is assuming similar neurovascular coupling; however, in the case of local ischemia or other pre-existing vascular disease (such as stenosis), this assumption may not be justified.[45] Such decoupling poses difficulties in interpreting the pathophysiological basis for differences between the groups (ie, neuronal or vascular). Alterations in the BOLD signal (eg, decreased amplitude) that result from mere vascular changes have been shown in patients with cerebrovascular disease.[46–48] Thus far, all studies examining the effect of changes in neurovascular coupling have used task-based fMRI. It is yet to be determined how such changes affect functional connectivity based on rs-fMRI. A multimodal approach including electroencephalography/magnetoencephalography as well as methods to assess cerebral blood flow quantitatively, such as ASL, may be used to separate neuronal from vascular changes. In addition, shifting from group comparisons of healthy controls and patients to longitudinal studies with single patient–based analysis could further minimize this limitation. Linking the observed changes in functional connectivity after stroke to relevant behavioral measures is another crucial factor, which would support neuronal bases for changes observed.

Changes in local perfusion and metabolism after stroke are most pronounced in the early acute phase (<24 hours after ictus). These complex hemodynamic changes are reflected in the BOLD signal because of dependency on cerebral blood flow, cerebral blood volume, and cerebral metabolic rate of oxygen.[8] Since the effects of hyperacute hemodynamic changes on functional connectivity rs-fMRI have not been explored thus far, results obtained at this stage should

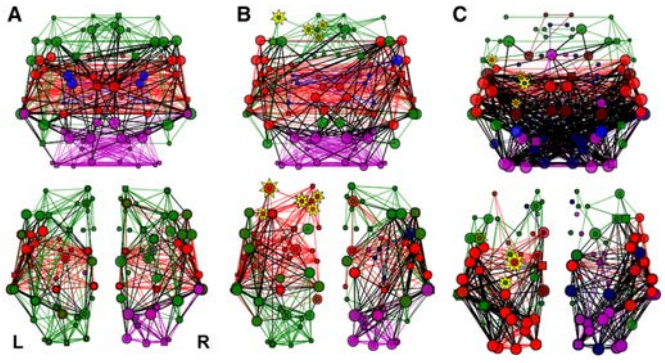

Figure 4. The topological role of the lesion has a crucial impact on the whole-brain network integrity. **A**, A healthy control template demonstrating an intact modular organization on the whole brain (**top**) and for each hemisphere separately (**bottom**). **B**, Patient with low connector damage demonstrating a relatively preserved modular organization. **C**, Patient with high connector damage demonstrating a highly disrupted modular organization at the whole-brain level as well as in both hemispheres. Yellow stars depict lesioned nodes, with the size of the star proportional to the percent damage to that node. Adapted from Gratton et al[39] with permission of the publisher. Copyright © 2012, The MIT Press. Authorization for this adaptation has been obtained both from the owner of the copyright in the original work and from the owner of copyright in the translation or adaptation.

be considered with caution. To minimize the effects of local perfusion changes, most studies explore patients after the first 24 hours and remove the lesion area from the analysis. These studies demonstrate that the changes in functional connectivity are not solely dependent on the lesion area.

Current methodological developments in the field of rs-fMRI are concerned with changes in functional connectivity during the rs-fMRI scan. All studies investigating functional connectivity in stroke published thus far have provided stationary information concerning the interaction between different brain regions. However, supported by findings demonstrating a link between different resting-state networks and specific combinations of electrophysiological rhythms,[49] recent studies have explored the dynamics of intrinsic BOLD fluctuations in healthy subjects as well as in schizophrenia, depression, and Alzheimer disease.[50] Such analysis requires longer scanning time, which could pose difficulties when applied to patients with stroke. Nonetheless, future studies exploring changes in dynamic connectivity may shed light on the underlying mechanisms of reported connectivity changes and behavioral deficits after stroke.

Based on the promising results obtained to date, more systematic validation studies in larger clinical populations with thorough clinical description are an important next step. This should allow for a validation of the multinetwork approach for an optimal description of neurological symptoms, as well as improved prognostic accuracy. The diagnostic assessment of connectivity changes in multiple networks finds a therapeutic counterpart in transcranial stimulation approaches, which have been shown to successfully modulate connectivity in cerebral networks.[2,3] In future studies, rs-fMRI connectivity patterns may be used to tailor stimulation protocol to individual patients.

Acknowledgments

We thank Dr Fatma Imamoglu, Dr Guido Hesselmann, Estrid Jakobsen, Alexander Schäfer, and Judy Kipping for their help and advice during the preparation of this article.

Sources of Funding

The study was supported by Minerva Stiftung, DFG (Berlin School of Mind and Brain), Einstein Foundation Berlin (The Mind-Brain Institute), BMBF (Center for Stroke Research Berlin), and the German Competence Net Stroke.

Disclosures

None.

References

1. Andrews RJ. Transhemispheric diaschisis. A review and comment. *Stroke*. 1991;22:943–949.
2. Grefkes C, Fink GR. Reorganization of cerebral networks after stroke: new insights from neuroimaging with connectivity approaches. *Brain*. 2011;134(pt 5):1264–1276.
3. Sehm B, Schäfer A, Kipping J, Margulies D, Conde V, Taubert M, et al. Dynamic modulation of intrinsic functional connectivity by transcranial direct current stimulation. *J Neurophysiol*. 2012;108:3253–3263.
4. Corbetta M. Functional connectivity and neurological recovery. *Dev Psychobiol*. 2012;54:239–253.
5. Biswal B, Yetkin FZ, Haughton VM, Hyde JS. Functional connectivity in the motor cortex of resting human brain using echo-planar MRI. *Magn Reson Med*. 1995;34:537–541.
6. Smith SM, Fox PT, Miller KL, Glahn DC, Fox PM, Mackay CE, et al. Correspondence of the brain's functional architecture during activation and rest. *Proc Natl Acad Sci U S A*. 2009;106:13040–13045.
7. Vincent JL, Patel GH, Fox MD, Snyder AZ, Baker JT, Van Essen DC, et al. Intrinsic functional architecture in the anaesthetized monkey brain. *Nature*. 2007;447:83–86.
8. Villringer A, Dirnagl U. Coupling of brain activity and cerebral blood flow: basis of functional neuroimaging. *Cerebrovasc Brain Metab Rev*. 1995;7:240–276.
9. Villringer A, Rosen BR, Belliveau JW, Ackerman JL, Lauffer RB, Buxton RB, et al. Dynamic imaging with lanthanide chelates in normal brain: contrast due to magnetic susceptibility effects. *Magn Reson Med*. 1988;6:164–174.
10. Koretsky AP. Early development of arterial spin labeling to measure regional brain blood flow by MRI. *Neuroimage*. 2012;62:602–607.
11. Petersen ET, Zimine I, Ho YC, Golay X. Non-invasive measurement of perfusion: a critical review of arterial spin labelling techniques. *Br J Radiol*. 2006;79:688–701.
12. Bokkers RP, Hernandez DA, Merino JG, Mirasol RV, van Osch MJ, Hendrikse J, et al; National Institutes of Health Stroke Natural History Investigators. Whole-brain arterial spin labeling perfusion MRI in patients with acute stroke. *Stroke*. 2012;43:1290–1294.
13. Lv Y, Margulies DS, Cameron Craddock R, Long X, Winter B, et al. Identifying the perfusion deficit in acute stroke with resting-state functional magnetic resonance imaging. *Ann Neurol*. 2013;73:136–140.
14. Amemiya S, Kunimatsu A, Saito N, Ohtomo K. Cerebral hemodynamic impairment: assessment with resting-state functional MR imaging. *Radiology*. 2014;270:548–555.
15. Feinberg DA, Moeller S, Smith SM, Auerbach E, Ramanna S, Gunther M, et al. Multiplexed echo planar imaging for sub-second whole brain FMRI and fast diffusion imaging. *PLoS One*. 2010;5:e15710.
16. Carmichael ST. Plasticity of cortical projections after stroke. *Neuroscientist*. 2003;9:64–75.
17. Stark DE, Margulies DS, Shehzad ZE, Reiss P, Kelly AM, Uddin LQ, et al. Regional variation in interhemispheric coordination of intrinsic hemodynamic fluctuations. *J Neurosci*. 2008;28:13754–13764.
18. Carter AR, Astafiev SV, Lang CE, Connor LT, Rengachary J, Strube MJ, et al. Resting interhemispheric functional magnetic resonance imaging connectivity predicts performance after stroke. *Ann Neurol*. 2010;67:365–375.
19. Carter AR, Patel KR, Astafiev SV, Snyder AZ, Rengachary J, Strube MJ, et al. Upstream dysfunction of somatomotor functional connectivity after corticospinal damage in stroke. *Neurorehabil Neural Repair*. 2012;26:7–19.
20. Park CH, Chang WH, Ohn SH, Kim ST, Bang OY, Pascual-Leone A, et al. Longitudinal changes of resting-state functional connectivity during motor recovery after stroke. *Stroke*. 2011;42:1357–1362.
21. Golestani AM, Tymchuk S, Demchuk A, Goodyear BG; VISION-2 Study Group. Longitudinal evaluation of resting-state FMRI after acute stroke with hemiparesis. *Neurorehabil Neural Repair*. 2013;27:153–163.
22. van Meer MP, van der Marel K, Wang K, Otte WM, El Bouazati S, Roeling TA, et al. Recovery of sensorimotor function after experimental stroke correlates with restoration of resting-state interhemispheric functional connectivity. *J Neurosci*. 2010;30:3964–3972.
23. van Meer MP, van der Marel K, Otte WM, Berkelbach van der Sprenkel JW, Dijkhuizen RM. Correspondence between altered functional and structural connectivity in the contralesional sensorimotor cortex after unilateral stroke in rats: a combined resting-state functional MRI and manganese-enhanced MRI study. *J Cereb Blood Flow Metab*. 2010;30:1707–1711.
24. He BJ, Snyder AZ, Vincent JL, Epstein A, Shulman GL, Corbetta M. Breakdown of functional connectivity in frontoparietal networks underlies behavioral deficits in spatial neglect. *Neuron*. 2007;53:905–918.
25. Gillebert CR, Mantini D, Thijs V, Sunaert S, Dupont P, Vandenberghe R. Lesion evidence for the critical role of the intraparietal sulcus in spatial attention. *Brain*. 2011;134(pt 6):1694–1709.
26. Buckner RL, Andrews-Hanna JR, Schacter DL. The brain's default network: anatomy, function, and relevance to disease. *Ann N Y Acad Sci*. 2008;1124:1–38.
27. Lassalle-Lagadec S, Sibon I, Dilharreguy B, Renou P, Fleury O, Allard M. Subacute default mode network dysfunction in the prediction of post-stroke depression severity. *Radiology*. 2012;264:218–224.
28. Tuladhar AM, Snaphaan L, Shumskaya E, Rijpkema M, Fernandez G, Norris DG, et al. Default Mode Network Connectivity in Stroke Patients. *PLoS One*. 2013;8:e66556.

29. Nomura EM, Gratton C, Visser RM, Kayser A, Perez F, D'Esposito M. Double dissociation of two cognitive control networks in patients with focal brain lesions. *Proc Natl Acad Sci U S A*. 2010;107:12017–12022.
30. Filippini N, MacIntosh BJ, Hough MG, Goodwin GM, Frisoni GB, Smith SM, et al. Distinct patterns of brain activity in young carriers of the APOE-epsilon4 allele. *Proc Natl Acad Sci U S A*. 2009;106:7209–7214.
31. Lohmann G, Ovadia-Caro S, Jungehülsing GJ, Margulies DS, Villringer A, Turner R. Connectivity concordance mapping: a new tool for model-free analysis of FMRI data of the human brain. *Front Syst Neurosci*. 2012;6:13.
32. Ovadia-Caro S, Villringer K, Fiebach J, Jungehulsing GJ, van der Meer E, Margulies DS, et al. Longitudinal effects of lesions on functional networks after stroke. *J Cereb Blood Flow Metab*. 2013;33:1279–1285.
33. Bullmore E, Sporns O. Complex brain networks: graph theoretical analysis of structural and functional systems. *Nat Rev Neurosci*. 2009;10:186–198.
34. Wang L, Yu C, Chen H, Qin W, He Y, Fan F, et al. Dynamic functional reorganization of the motor execution network after stroke. *Brain*. 2010;133(pt 4):1224–1238.
35. van Meer MP, Otte WM, van der Marel K, Nijboer CH, Kavelaars A, van der Sprenkel JW, et al. Extent of bilateral neuronal network reorganization and functional recovery in relation to stroke severity. *J Neurosci*. 2012;32:4495–4507.
36. Varoquaux G, Craddock RC. Learning and comparing functional connectomes across subjects. *Neuroimage*. 2013;80:405–415.
37. Honey CJ, Sporns O. Dynamical consequences of lesions in cortical networks. *Hum Brain Mapp*. 2008;29:802–809.
38. Alstott J, Breakspear M, Hagmann P, Cammoun L, Sporns O. Modeling the impact of lesions in the human brain. *PLoS Comput Biol*. 2009;5:e1000408.
39. Gratton C, Nomura EM, Pérez F, D'Esposito M. Focal brain lesions to critical locations cause widespread disruption of the modular organization of the brain. *J Cogn Neurosci*. 2012;24:1275–1285.
40. Power JD, Barnes KA, Snyder AZ, Schlaggar BL, Petersen SE. Spurious but systematic correlations in functional connectivity MRI networks arise from subject motion. *Neuroimage*. 2012;59:2142–2154.
41. Damoiseaux JS, Greicius MD. Greater than the sum of its parts: a review of studies combining structural connectivity and resting-state functional connectivity. *Brain Struct Funct*. 2009;213:525–533.
42. Schaefer A, Quinque E, Kipping J, Arélin K, Frisch S, Roggenhofer E, et al. Early small vessel disease affects frontoparietal and cerebellar hubs in close correlation with clinical symptoms – a resting-state fmri study [published online ahead of print April 30, 2014]. *J Cerebr Blood Flow Met*. doi:10.1038/jcbfm.2014.70
43. Johansen-Berg H, Scholz J, Stagg CJ. Relevance of structural brain connectivity to learning and recovery from stroke. *Front Syst Neurosci*. 2010;4:146.
44. Logothetis NK. The underpinnings of the BOLD functional magnetic resonance imaging signal. *J Neurosci*. 2003;23:3963–3971.
45. D'Esposito M, Deouell LY, Gazzaley A. Alterations in the BOLD fMRI signal with ageing and disease: a challenge for neuroimaging. *Nat Rev Neurosci*. 2003;4:863–872.
46. Pineiro R, Pendlebury S, Johansen-Berg H, Matthews PM. Altered hemodynamic responses in patients after subcortical stroke measured by functional MRI. *Stroke*. 2002;33:103–109.
47. Krainik A, Hund-Georgiadis M, Zysset S, von Cramon DY. Regional impairment of cerebrovascular reactivity and BOLD signal in adults after stroke. *Stroke*. 2005;36:1146–1152.
48. Mazzetto-Betti KC, Leoni RF, Pontes-Neto OM, Santos AC, Leite JP, Silva AC, et al. The stability of the blood oxygenation level-dependent functional MRI response to motor tasks is altered in patients with chronic ischemic stroke. *Stroke*. 2010;41:1921–1926.
49. Mantini D, Perrucci MG, Del Gratta C, Romani GL, Corbetta M. Electrophysiological signatures of resting state networks in the human brain. *Proc Natl Acad Sci U S A*. 2007;104:13170–13175.
50. Hutchison RM, Womelsdorf T, Allen EA, Bandettini PA, Calhoun VD, Corbetta M, et al. Dynamic functional connectivity: promise, issues, and interpretations. *Neuroimage*. 2013;80:360–378.

KEY WORDS: fMRI ■ stroke

SUPPLEMENTAL MATERIAL

Supplementary Table I: Summary of stroke studies using resting-state fMRI

Author	Studied network	Acq. Post-stroke	Lesion location and etiology	Analysis type	Main objective	Main findings
Lv et al., 2013[1]	Whole-brain analysis	1 day post-stroke	Heterogeneous locations Ischemic stroke	Time-shift analysis (correlating individual voxels with the global signal in different delays)	Investigate the time delay in the BOLD signal on the voxel level, and look for surrogate measures of the hypoperfused area	Time-shift analysis using rs-fMRI can provide information comparable to perfusion imaging and concerning areas of compromised perfusion, even in the acute phase
Amemiya et al., 2013[2]	Whole-brain analysis	1 day post-stroke and patients with chronic stenoocclusive vessel disease	Heterogeneous locations Ischemic stroke and MCA stenoocclusive vessel disease	Time-shift analysis	Test the ability to assess cerebral hemodynamic impairments using non invasive rs-fMRI	High correspondence between areas demonstrating a time delay and areas of hypoperfusion in both acute and chronic patients
Carter et al., 2010[3]	Arm subregion of sensorimotor network and dorsal attention network	≤ 4 weeks post-stroke	Cortical and subcortical lesions. Ischemic stroke (n=18) Hemorrhagic stroke (n=5)	Inter-hemispheric and intra-hemispheric seed-based FC analysis (seeds were defined based on previous task-based fMRI study in healthy controls)	Examine the link between behavioral performance and FC strength in attention and motor networks	Inter-hemispheric connectivity found to correlate with corresponding behavioral deficits. Intra-hemispheric connectivity did not demonstrate this relationship
Carter et al., 2012[4]	Arm subregion of sensorimotor network	≤ 4 weeks post-stroke	Corticospinal damage (cortical and subcortical) NM	Inter-hemispheric and intra-hemispheric seed-based FC analysis	Examine the link between extent of corticospinal damage and changes in cerebral FC	Corticospinal damage was negatively correlated with inter-hemispheric FC. This relationship was not found for intra-hemispheric FC

Author	Studied network	Acq. Post-stroke	Lesion location and etiology	Analysis type	Main objective	Main findings
Park et al., 2011[5]	Motor network	< 2 weeks, and 1, 3 and 6 months post-stroke	Supratentorial lesions. MCA (n=8), CR (n=2), ACA (n=1), SC (n=1) Ischemic stroke	FC from the ipsilesional primary motor cortex	Examine longitudinal changes in FC patterns	Increased connectivity with ipsilesional regions and decreased connectivity with contralesional regions as compared to controls. FC at onset correlated with motor recovery at 6 months post-stroke
Golestani et al., 2013[6]	Sensorimotor network	< 24 hours, 7 and 90 days post-stroke	Cortical (n=11), subcortical/WM (n=17), cerebellar/brain stem (n=3) Ischemic stroke	FC from the ipsilesional primary sensorimotor cortex and Inter-hemispheric FC based on the same seed	Examine longitudinal changes in FC patterns	Decreased inter-hemispheric connectivity in the acute phase only in patients with motor deficits. Connectivity recovered 7 days post-stroke in recovered patients. 90 days post-stroke inter-hemispheric connectivity recovered, yet decreased connectivity with subcortical regions remained
van Meer et al., 2010[7]	Sensorimotor network	2 days pre-surgery and 3,7,21 and 70 days post-surgery (experimental stroke)	Occlusion of the right MCA. Ischemic stroke	Inter-hemispheric and intra-hemispheric FC for the cortical sensorimotor network (the forelimb region of the primary somatosensory cortex and primary motor cortex)	Study changes in FC within the bilateral sensorimotor network and to link these changes to behavioral deficits	Decreased inter-hemispheric connectivity combined with impaired behavioral performance early after stroke. Recovery of inter-hemispheric connectivity was associated with improved behavioral performance. Increase in intra-hemispheric connectivity was found in animals with large strokes.

Author	Studied network	Acq. Post-stroke	Lesion location and etiology	Analysis type	Main objective	Main findings
van Meer et al., 2010[8]	Sensorimotor network	10 weeks post-surgery (experimental stroke)	Occlusion of the right MCA. Ischemic stroke	FC from the primary motor cortex, inter-hemispheric and intra-hemispheric connectivity for sensorimotor network	Examine the link between alterations in FC following stroke and neuroanatomical connectivity as measured by manganese-enhanced MRI	A decrease in inter-hemispheric connectivity was associated with decrease in transcallosal tracer transfer, while increased intra-hemispheric connectivity was associated with a local increase in tracer uptake
He et al., 2007[9]	Attention network	30 days (30±23), and 40 weeks (40±11) post-stroke	Right frontoparietal stroke (cortical and subcortical). Ischemic and Hemorrhagic stroke	FC and inter-hemispheric connectivity based on 8 regions in the dorsal attention network and 5 regions in the ventral attention network	Examine longitudinal changes in FC for two attention networks and the link between FC changes and behavioral impairment	Decreased inter-hemispheric connectivity for both dorsal and ventral attention networks. In the dorsal network, connectivity recovered at the chronic stage. Decreased connectivity correlated with behavioral impairment.
Gillebert et al., 2011[10]	Attention network	107 days post-stroke	Intra-parietal sulcus lesions. Ischemic stroke	Independent component analysis (used spatial correlation with maps obtained from controls)	Examine the influence of intra-parietal lesions on FC of the attention network	Decreased spatial correlation specific to the relevant network involved (depending on the lesion location)

Author	Studied network	Acq. Post-stroke	Lesion location and etiology	Analysis type	Main objective	Main findings
Lassalle-Lagadec et al., 2012[11]	Default-mode network (DMN)	10 days post-stroke	Heterogeneous locations sparing the DMN -Occipital cortex (N=3), Frontal cortex (N=1), Insular cortex (N=4), subcortical (N=16). Ischemic stroke	Independent component analysis	Link changes in FC within the DMN with depression and anxiety symptoms	Abnormal DMN connectivity at 10 days post-stroke was correlated with depression scores obtained 3 months post-stroke. Abnormal DMN connectivity in various structures was correlated with both early and late anxiety severity
Tuladhar et al., 2013[12]	Default-mode network	9-12 weeks post stroke	IC (N=5), CR (N=4), thalamus (N=2), Occipital lobe (N=4), brainstem (N=4), Parietal lobe (N=1). Ischemic stroke	Independent component analysis	Link changes in FC within the DMN with episodic memory dysfunction	Decreased FC within DMN in stroke patients as compared to controls. Correlation between FC and behavior was found only for controls
Nomura et al., 2010[13]	Fronto-parietal and Cingulo-opercular networks	At least 5 months post-stroke	Heterogeneous locations (cortical and subcortical). Ischemic stroke (N=16) Hemorrhagic stroke (N=1) Tumor resection (N=2), TBI (N=2)	FC based on mean correlation within and between network nodes, graph theory measures based on the pre-defined regions	Demonstrating dissociation of two networks based on changes in FC with heterogeneous lesions	The degree of decreased connectivity correlated with the degree of network damage only in the affected network. Intact nodes within the damaged network demonstrate impaired graph measures as compared to nodes in the unaffected network

Author	Studied network	Acq. Post-stroke	Lesion location and etiology	Analysis type	Main objective	Main findings
Ovadia-Caro et al., 2013[14]	Eight pre-defined networks (whole-brain analysis)	1,7, and 90 days post-stroke	Heterogeneous locations (cortical and subcortical), no brainstem, no cerebellar lesions. Ischemic stroke	Dual regression analysis and spatial concordance of FC maps	Investigate the generalizability of dissociation between different networks on the whole-brain level, and provide a methodological approach for heterogeneous locations of lesions	Over time, changes in FC were more pronounced in affected networks as compared to unaffected networks. Change in connectivity correlated with behavioral change over time
Wang et al., 2010[15]	Motor network	1 week, 2 weeks, 1 month, 3 months and 1 year post-stroke	Subcortical strokes - IC involvement (N=10/10), CR involvement (N=8/10), BG involvement (N=4/10). NM	Graph theory measures As well as FC between the nodes (21 regions)	Investigate longitudinal changes in the topology of the motor execution network	A gradual shift in the network topology to a random mode. Alterations in regional centrality and FC. A gradual increase in FC between ipsilesional motor cortex and contralesional motor areas. A correlation between connectivity measures and behavioral trajectory
van Meer et al., 2012[16]	Sensorimotor network	2 days pre-surgery, and 3,7,21,49, 70 days post-surgery (experimental stroke)	Occlusion of the right MCA. Ischemic stroke	Graph theory measures and FC	Link longitudinal changes in functional and structural organization to changes in sensorimotor function	A gradual recovery of network topology, along with normalization of inter-hemispheric FC. Correlation between connectivity changes and behavioral improvement

Author	Studied network	Acq. Post-stroke	Lesion location and etiology	Analysis type	Main objective	Main findings
Gratton et al., 2012[17]	Whole-brain analysis	Chronic (mean 7y. post-stroke, minimum 2 months post-stroke)	Heterogeneous locations (mainly cortical). Stroke (N=25), TBI (N=6), tumors (N=4)	Graph theory measures (based on the AAL template)	Test the influence of differences in the topological role of lesions on the integrity of whole-network connectivity	Decrease in modularity correlated with the increase in damage to connector hubs and not to provincial hubs. Modularity impaired in both affected and unaffected hemispheres

Abbreviations: Acq = Acquisition time, BOLD = blood oxygenation level-dependent, rs-fMRI = resting-state functional magnetic resonance imaging, MCA = middle cerebral artery, FC = functional connectivity, CR = corona radiate, ACA = anterior cerebral artery, SC = striatocapsular, WM = white matter, DMN = default-mode network, IC = internal capsule, BG = basal ganglia, TBI = traumatic brain injury, AAL = automated anatomical labeling, NM = not mentioned.

Supplemental References:

1. Lv Y, Margulies DS, Cameron Craddock R, Long X, Winter B, et al. Identifying the perfusion deficit in acute stroke with resting-state functional magnetic resonance imaging. Ann Neurol. 2013;73:136-140
2. Amemiya S, Kunimatsu A, Saito N, Ohtomo K. Cerebral hemodynamic impairment: Assessment with resting-state functional mr imaging. Radiology. 2013;270:548-555
3. Carter AR, Astafiev SV, Lang CE, Connor LT, Rengachary J, Strube MJ, et al. Resting interhemispheric functional magnetic resonance imaging connectivity predicts performance after stroke. Ann Neurol. 2010;67:365-375
4. Carter AR, Patel KR, Astafiev SV, Snyder AZ, Rengachary J, Strube MJ, et al. Upstream dysfunction of somatomotor functional connectivity after corticospinal damage in stroke. Neurorehabil Neural Repair. 2012;26:7-19
5. Park CH, Chang WH, Ohn SH, Kim ST, Bang OY, Pascual-Leone A, et al. Longitudinal changes of resting-state functional connectivity during motor recovery after stroke. Stroke. 2011;42:1357-1362
6. Golestani AM, Tymchuk S, Demchuk A, Goodyear BG. Longitudinal evaluation of resting-state fmri after acute stroke with hemiparesis. Neurorehabil Neural Repair. 2013;27:153-163
7. van Meer MP, van der Marel K, Wang K, Otte WM, El Bouazati S, Roeling TA, et al. Recovery of sensorimotor function after experimental stroke correlates with restoration of resting-state interhemispheric functional connectivity. J Neurosci. 2010;30:3964-3972
8. van Meer MP, van der Marel K, Otte WM, Berkelbach van der Sprenkel JW, Dijkhuizen RM. Correspondence between altered functional and structural connectivity in the contralesional sensorimotor cortex after unilateral stroke in rats: A combined resting-state functional mri and manganese-enhanced mri study. J Cereb Blood Flow Metab. 2010;30:1707-1711
9. He BJ, Snyder AZ, Vincent JL, Epstein A, Shulman GL, Corbetta M. Breakdown of functional connectivity in frontoparietal networks underlies behavioral deficits in spatial neglect. Neuron. 2007;53:905-918
10. Gillebert CR, Mantini D, Thijs V, Sunaert S, Dupont P, Vandenberghe R. Lesion evidence for the critical role of the intraparietal sulcus in spatial attention. Brain. 2011;134:1694-1709
11. Lassalle-Lagadec S, Sibon I, Dilharreguy B, Renou P, Fleury O, Allard M. Subacute default mode network dysfunction in the prediction of post-stroke depression severity. Radiology. 2012;264:218-224
12. Tuladhar AM, Snaphaan L, Shumskaya E, Rijpkema M, Fernandez G, Norris DG, et al. Default mode network connectivity in stroke patients. PLoS ONE. 2013;8:e66556
13. Nomura EM, Gratton C, Visser RM, Kayser A, Perez F, D'Esposito M. Double dissociation of two cognitive control networks in patients with focal brain lesions. Proc Natl Acad Sci U S A. 2010;107:12017-12022
14. Ovadia-Caro S, Villringer K, Fiebach J, Jungehulsing GJ, van der Meer E, Margulies DS, et al. Longitudinal effects of lesions on functional networks after stroke. J Cereb Blood Flow Metab. 2013;33:1279-1285

15. Wang L, Yu C, Chen H, Qin W, He Y, Fan F, et al. Dynamic functional reorganization of the motor execution network after stroke. Brain. 2010;133:1224-1238
16. van Meer MP, Otte WM, van der Marel K, Nijboer CH, Kavelaars A, van der Sprenkel JW, et al. Extent of bilateral neuronal network reorganization and functional recovery in relation to stroke severity. J Neurosci. 2012;32:4495-4507
17. Gratton C, Nomura EM, Perez F, D'Esposito M. Focal brain lesions to critical locations cause widespread disruption of the modular organization of the brain. J Cogn Neurosci. 2012;24:1275-1285

The Value of Resting-State Functional Magnetic Resonance Imaging in Stroke

Smadar Ovadia-Caro, Daniel S. Margulies and Arno Villringer

Stroke. 2014;45:2818-2824; originally published online July 10, 2014;
doi: 10.1161/STROKEAHA.114.003689
Stroke is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231

Print ISSN: 0039-2499. Online ISSN: 1524-4628

MPI Series in Human Cognitive and Brain Sciences:

1 Anja Hahne
Charakteristika syntaktischer und semantischer Prozesse bei der auditiven Sprachverarbeitung: Evidenz aus ereigniskorrelierten Potentialstudien

2 Ricarda Schubotz
Erinnern kurzer Zeitdauern: Behaviorale und neurophysiologische Korrelate einer Arbeitsgedächtnisfunktion

3 Volker Bosch
Das Halten von Information im Arbeitsgedächtnis: Dissoziationen langsamer corticaler Potentiale

4 Jorge Jovicich
An investigation of the use of Gradient- and Spin-Echo (GRASE) imaging for functional MRI of the human brain

5 Rosemary C. Dymond
Spatial Specificity and Temporal Accuracy in Functional Magnetic Resonance Investigations

6 Stefan Zysset
Eine experimentalpsychologische Studie zu Gedächtnisabrufprozessen unter Verwendung der funktionellen Magnetresonanztomographie

7 Ulrich Hartmann
Ein mechanisches Finite-Elemente-Modell des menschlichen Kopfes

8 Bertram Opitz
Funktionelle Neuroanatomie der Verarbeitung einfacher und komplexer akustischer Reize: Integration haemodynamischer und elektrophysiologischer Maße

9 Gisela Müller-Plath
Formale Modellierung visueller Suchstrategien mit Anwendungen bei der Lokalisation von Hirnfunktionen und in der Diagnostik von Aufmerksamkeitsstörungen

10 Thomas Jacobsen
Characteristics of processing morphological structural and inherent case in language comprehension

11 Stefan Kölsch
Brain and Music
A contribution to the investigation of central auditory processing with a new electrophysiological approach

12 Stefan Frisch
Verb-Argument-Struktur, Kasus und thematische Interpretation beim Sprachverstehen

13 Markus Ullsperger
The role of retrieval inhibition in directed forgetting – an event-related brain potential analysis

14 Martin Koch
Measurement of the Self-Diffusion Tensor of Water in the Human Brain

15 Axel Hutt
Methoden zur Untersuchung der Dynamik raumzeitlicher Signale

16 Frithjof Kruggel
Detektion und Quantifizierung von Hirnaktivität mit der funktionellen Magnetresonanztomographie

17 Anja Dove
Lokalisierung an internen Kontrollprozessen beteiligter Hirngebiete mithilfe des Aufgabenwechselparadigmas und der ereigniskorrelierten funktionellen Magnetresonanztomographie

18 Karsten Steinhauer
Hirnphysiologische Korrelate prosodischer Satzverarbeitung bei gesprochener und geschriebener Sprache

19 Silke Urban
Verbinformationen im Satzverstehen

20 Katja Werheid
Implizites Sequenzlernen bei Morbus Parkinson

21 Doreen Nessler
Is it Memory or Illusion? Electrophysiological Characteristics of True and False Recognition

22 Christoph Herrmann
Die Bedeutung von 40-Hz-Oszillationen für kognitive Prozesse

23 Christian Fiebach
Working Memory and Syntax during Sentence Processing.
A neurocognitive investigation with event-related brain potentials and functional magnetic resonance imaging

24 Grit Hein
Lokalisation von Doppelaufgabendefiziten bei gesunden älteren Personen und neurologischen Patienten

25 Monica de Filippis
Die visuelle Verarbeitung unbeachteter Wörter. Ein elektrophysiologischer Ansatz

26 Ulrich Müller
Die katecholaminerge Modulation präfrontaler kognitiver Funktionen beim Menschen

27 Kristina Uhl
Kontrollfunktion des Arbeitsgedächtnisses über interferierende Information

28 Ina Bornkessel
The Argument Dependency Model: A Neurocognitive Approach to Incremental Interpretation

29 Sonja Lattner
Neurophysiologische Untersuchungen zur auditorischen Verarbeitung von Stimminformationen

30 Christin Grünewald
Die Rolle motorischer Schemata bei der Objektrepräsentation: Untersuchungen mit funktioneller Magnetresonanztomographie

31 Annett Schirmer
Emotional Speech Perception: Electrophysiological Insights into the Processing of Emotional Prosody and Word Valence in Men and Women

32 André J. Szameitat
Die Funktionalität des lateral-präfrontalen Cortex für die Verarbeitung von Doppelaufgaben

33 Susanne Wagner
Verbales Arbeitsgedächtnis und die Verarbeitung ambiger Wörter in Wort- und Satzkontexten

34 Sophie Manthey
Hirn und Handlung: Untersuchung der Handlungsrepräsentation im ventralen prämotorischen Cortex mit Hilfe der funktionellen Magnet-Resonanz-Tomographie

35 Stefan Heim
Towards a Common Neural Network Model of Language Production and Comprehension: fMRI Evidence for the Processing of Phonological and Syntactic Information in Single Words

36 Claudia Friedrich
Prosody and spoken word recognition: Behavioral and ERP correlates

37 Ulrike Lex
Sprachlateralisierung bei Rechts- und Linkshändern mit funktioneller Magnetresonanztomographie

38 Thomas Arnold
Computergestützte Befundung klinischer Elektroenzephalogramme

39 Carsten H. Wolters
Influence of Tissue Conductivity Inhomogeneity and Anisotropy on EEG/MEG based Source Localization in the Human Brain

40 Ansgar Hantsch
Fisch oder Karpfen? Lexikale Aktivierung von Benennungsalternative bei der Objektbenennung

41 Peggy Bungert
Zentralnervöse Verarbeitung akustischer Informationen Signalidentifikation, Signallateralisation und zeitgebundene Informationsverarbeitung bei Patienten mit erworbenen Hirnschädigungen

42 Daniel Senkowski
Neuronal correlates of selective attention: An investigation of electrophysiological brain responses in the EEG and MEG

43 Gert Wollny
Analysis of Changes in Temporal Series of Medical Images

S 1 Markus Ullsperger & Michael Falkenstein
Errors, Conflicts, and the Brain Current Opinions on Performance Monitoring

44 Angelika Wolf
Sprachverstehen mit Cochlea-Implantat: EKP-Studien mit postlingual ertaubten erwachsenen CI-Trägern

45 Kirsten G. Volz
Brain correlates of uncertain decisions: Types and degrees of uncertainty

46 Hagen Huttner
Magnetresonanztomographische Untersuchungen über die anatomische Variabilität des Frontallappens des menschlichen Großhirns

47 Dirk Köster
Morphology and Spoken Word Comprehension: Electrophysiological Investigations of Internal Compound Structure

48 Claudia A. Hruska
Einflüsse kontextueller und prosodischer Informationen in der auditorischen Satzverarbeitung: Untersuchungen mit ereigniskorrelierten Hirnpotentialen

49 Hannes Ruge
Eine Analyse des raum-zeitlichen Musters neuronaler Aktivierung im Aufgabenwechselparadigma zur Untersuchung handlungssteuernder Prozesse

50 Ricarda I. Schubotz
Human premotor cortex: Beyond motor performance

51 Clemens von Zerssen
Bewusstes Erinnern und falsches Wiedererkennen: Eine funktionelle MRT Studie neuroanatomischer Gedächtniskorrelate

52 Christiane Weber
Rhythm is gonna get you.
Electrophysiological markers of rhythmic processing in infants with and without risk for Specific Language Impairment (SLI)

53 Marc Schönwiesner
Functional Mapping of Basic Acoustic Parameters in the Human Central Auditory System

54 Katja Fiehler
Temporospatial characteristics of error correction

55 Britta Stolterfoht
Processing Word Order Variations and Ellipses: The Interplay of Syntax and Information Structure during Sentence Comprehension

56 Claudia Danielmeier
Neuronale Grundlagen der Interferenz zwischen Handlung und visueller Wahrnehmung

57 Margret Hund-Georgiadis
Die Organisation von Sprache und ihre Reorganisation bei ausgewählten, neurologischen Erkrankungen gemessen mit funktioneller Magnetresonanztomographie – Einflüsse von Händigkeit, Läsion, Performanz und Perfusion

58 Jutta L. Mueller
Mechanisms of auditory sentence comprehension in first and second language: An electrophysiological miniature grammar study

59 Franziska Biedermann
Auditorische Diskriminationsleistungen nach unilateralen Läsionen im Di- und Telenzephalon

60 Shirley-Ann Rüschemeyer
The Processing of Lexical Semantic and Syntactic Information in Spoken Sentences: Neuroimaging and Behavioral Studies of Native and Non-Native Speakers

61 Kerstin Leuckefeld
The Development of Argument Processing Mechanisms in German. An Electrophysiological Investigation with School-Aged Children and Adults

62 Axel Christian Kühn
Bestimmung der Lateralisierung von Sprachprozessen unter besondere Berücksichtigung des temporalen Cortex, gemessen mit fMRT

63 Ann Pannekamp
Prosodische Informationsverarbeitung bei normalsprachlichem und deviantem Satzmaterial: Untersuchungen mit ereigniskorrelierten Hirnpotentialen

64 Jan Derrfuß
Functional specialization in the lateral frontal cortex: The role of the inferior frontal junction in cognitive control

65 Andrea Mona Philipp
The cognitive representation of tasks – Exploring the role of response modalities using the task-switching paradigm

66 Ulrike Toepel
Contrastive Topic and Focus Information in Discourse – Prosodic Realisation and Electrophysiological Brain Correlates

67 Karsten Müller
Die Anwendung von Spektral- und Waveletanalyse zur Untersuchung der Dynamik von BOLD-Zeitreihen verschiedener Hirnareale

68 Sonja A.Kotz
The role of the basal ganglia in auditory language processing: Evidence from ERP lesion studies and functional neuroimaging

69 Sonja Rossi
The role of proficiency in syntactic second language processing: Evidence from event-related brain potentials in German and Italian

70 Birte U. Forstmann
Behavioral and neural correlates of endogenous control processes in task switching

71 Silke Paulmann
Electrophysiological Evidence on the Processing of Emotional Prosody: Insights from Healthy and Patient Populations

72 Matthias L. Schroeter
Enlightening the Brain – Optical Imaging in Cognitive Neuroscience

73 Julia Reinholz
Interhemispheric interaction in object- and word-related visual areas

74 Evelyn C. Ferstl
The Functional Neuroanatomy of Text Comprehension

75 Miriam Gade
Aufgabeninhibition als Mechanismus der Konfliktreduktion zwischen Aufgabenrepräsentationen

76 Juliane Hofmann
Phonological, Morphological, and Semantic Aspects of Grammatical Gender Processing in German

77 Petra Augurzky
Attaching Relative Clauses in German – The Role of Implicit and Explicit Prosody in Sentence Processing

78 Uta Wolfensteller
Habituelle und arbiträre sensomotorische Verknüpfungen im lateralen prämotorischen Kortex des Menschen

79 Päivi Sivonen
Event-related brain activation in speech perception: From sensory to cognitive processes

80 Yun Nan
Music phrase structure perception: the neural basis, the effects of acculturation and musical training

81 Katrin Schulze
Neural Correlates of Working Memory for Verbal and Tonal Stimuli in Nonmusicians and Musicians With and Without Absolute Pitch

82 Korinna Eckstein
Interaktion von Syntax und Prosodie beim Sprachverstehen: Untersuchungen anhand ereigniskorrelierter Hirnpotentiale

83 Florian Th. Siebörger
Funktionelle Neuroanatomie des Textverstehens: Kohärenzbildung bei Witzen und anderen ungewöhnlichen Texten

84 Diana Böttger
Aktivität im Gamma-Frequenzbereich des EEG: Einfluss demographischer Faktoren und kognitiver Korrelate

85 Jörg Bahlmann
Neural correlates of the processing of linear and hierarchical artificial grammar rules: Electrophysiological and neuroimaging studies

86 Jan Zwickel
Specific Interference Effects Between Temporally Overlapping Action and Perception

87 Markus Ullsperger
Functional Neuroanatomy of Performance Monitoring: fMRI, ERP, and Patient Studies

88 Susanne Dietrich
Vom Brüllen zum Wort – MRT-Studien zur kognitiven Verarbeitung emotionaler Vokalisationen

89 Maren Schmidt-Kassow
What's Beat got to do with ist? The Influence of Meter on Syntactic Processing: ERP Evidence from Healthy and Patient populations

90 Monika Lück
Die Verarbeitung morphologisch komplexer Wörter bei Kindern im Schulalter: Neurophysiologische Korrelate der Entwicklung

91 Diana P. Szameitat
Perzeption und akustische Eigenschaften von Emotionen in menschlichem Lachen

92 Beate Sabisch
Mechanisms of auditory sentence comprehension in children with specific language impairment and children with developmental dyslexia: A neurophysiological investigation

93 Regine Oberecker
Grammatikverarbeitung im Kindesalter: EKP-Studien zum auditorischen Satzverstehen

94 Şükrü Barış Demiral
Incremental Argument Interpretation in Turkish Sentence Comprehension

95 Henning Holle
The Comprehension of Co-Speech Iconic Gestures: Behavioral, Electrophysiological and Neuroimaging Studies

96 Marcel Braß
Das inferior frontale Kreuzungsareal und seine Rolle bei der kognitiven Kontrolle unseres Verhaltens

97 Anna S. Hasting
Syntax in a blink: Early and automatic processing of syntactic rules as revealed by event-related brain potentials

98 Sebastian Jentschke
Neural Correlates of Processing Syntax in Music and Language – Influences of Development, Musical Training and Language Impairment

99 Amelie Mahlstedt
The Acquisition of Case marking Information as a Cue to Argument Interpretation in German
An Electrophysiological Investigation with Pre-school Children

100 Nikolaus Steinbeis
Investigating the meaning of music using EEG and fMRI

101 Tilmann A. Klein
Learning from errors: Genetic evidence for a central role of dopamine in human performance monitoring

102 Franziska Maria Korb
Die funktionelle Spezialisierung des lateralen präfrontalen Cortex: Untersuchungen mittels funktioneller Magnetresonanztomographie

103 Sonja Fleischhauer
Neuronale Verarbeitung emotionaler Prosodie und Syntax: die Rolle des verbalen Arbeitsgedächtnisses

104 Friederike Sophie Haupt
The component mapping problem: An investigation of grammatical function reanalysis in differing experimental contexts using eventrelated brain potentials

105 Jens Brauer
Functional development and structural maturation in the brain's neural network underlying language comprehension

106 Philipp Kanske
Exploring executive attention in emotion: ERP and fMRI evidence

107 Julia Grieser Painter
Music, meaning, and a semantic space for musical sounds

108 Daniela Sammler
The Neuroanatomical Overlap of Syntax Processing in Music and Language - Evidence from Lesion and Intracranial ERP Studies

109 Norbert Zmyj
Selective Imitation in One-Year-Olds: How a Model's Characteristics Influence Imitation

110 Thomas Fritz
Emotion investigated with music of variable valence – neurophysiology and cultural influence

111 Stefanie Regel
The comprehension of figurative language: Electrophysiological evidence on the processing of irony

112 Miriam Beisert
Transformation Rules in Tool Use

113 Veronika Krieghoff
Neural correlates of Intentional Actions

114 Andreja Bubić
Violation of expectations in sequence processing

115 Claudia Männel
Prosodic processing during language acquisition: Electrophysiological studies on intonational phrase processing

116 Konstanze Albrecht
Brain correlates of cognitive processes underlying intertemporal choice for self and other

117 Katrin Sakreida
Nicht-motorische Funktionen des prämotorischen Kortex: Patientenstudien und funktionelle Bildgebung

118 Susann Wolff
The interplay of free word order and pro-drop in incremental sentence processing: Neurophysiological evidence from Japanese

119 Tim Raettig
The Cortical Infrastructure of Language Processing: Evidence from Functional and Anatomical Neuroimaging

120 Maria Golde
Premotor cortex contributions to abstract and action-related relational processing

121 Daniel S. Margulies
Resting-State Functional Connectivity fMRI: A new approach for assessing functional neuroanatomy in humans with applications to neuroanatomical, developmental and clinical questions

122 Franziska Süß
The interplay between attention and syntactic processes in the adult and developing brain: ERP evidences

123 Stefan Bode
From stimuli to motor responses: Decoding rules and decision mechanisms in the human brain

124 Christiane Diefenbach
Interactions between sentence comprehension and concurrent action: The role of movement effects and timing

125 Moritz M. Daum
Mechanismen der frühkindlichen Entwicklung des Handlungsverständnisses

126 Jürgen Dukart
Contribution of FDG-PET and MRI to improve Understanding, Detection and Differentiation of Dementia

127 Kamal Kumar Choudhary
Incremental Argument Interpretation in a Split Ergative Language: Neurophysiological Evidence from Hindi

128 Peggy Sparenberg
Filling the Gap: Temporal and Motor Aspects of the Mental Simulation of Occluded Actions

129 Luming Wang
The Influence of Animacy and Context on Word Order Processing: Neurophysiological Evidence from Mandarin Chinese

130 Barbara Ettrich
Beeinträchtigung frontomedianer Funktionen bei Schädel-Hirn-Trauma

131 Sandra Dietrich
Coordination of Unimanual Continuous Movements with External Events

132 R. Muralikrishnan
An Electrophysiological Investigation Of Tamil Dative-Subject Constructions

133 Christian Obermeier
Exploring the significance of task, timing and background noise on gesture-speech integration

134 Björn Herrmann
Grammar and perception: Dissociation of early auditory processes in the brain

135 Eugenia Solano-Castiella
In vivo anatomical segmentation of the human amygdala and parcellation of emotional processing

136 Marco Taubert
Plastizität im sensomotorischen System – Lerninduzierte Veränderungen in der Struktur und Funktion des menschlichen Gehirns

137 Patricia Garrido Vásquez
Emotion Processing in Parkinson's Disease: The Role of Motor Symptom Asymmetry

138 Michael Schwartze
Adaptation to temporal structure

139 Christine S. Schipke
Processing Mechanisms of Argument Structure and Case-marking in Child Development: Neural Correlates and Behavioral Evidence

140 Sarah Jessen
Emotion Perception in the Multisensory Brain

141 Jane Neumann
Beyond activation detection: Advancing computational techniques for the analysis of functional MRI data

142 Franziska Knolle
Knowing what's next: The role of the cerebellum in generating predictions

143 Michael Skeide
Syntax and semantics networks in the developing brain

144 Sarah M. E. Gierhan
Brain networks for language
Anatomy and functional roles of neural pathways supporting language comprehension and repetition

145 Lars Meyer
The Working Memory of Argument-Verb Dependencies
Spatiotemporal Brain Dynamics during Sentence Processing

146 Benjamin Stahl
Treatment of Non-Fluent Aphasia through Melody, Rhythm and Formulaic Language

147 Kathrin Rothermich
The rhythm's gonna get you: ERP and fMRI evidence on the interaction of metric and semantic processing

148 Julia Merrill
Song and Speech Perception – Evidence from fMRI, Lesion Studies and Musical Disorder

149 Klaus-Martin Krönke
Learning by Doing?
Gesture-Based Word-Learning and its Neural Correlates in Healthy Volunteers and Patients with Residual Aphasia

150 Lisa Joana Knoll
When the hedgehog kisses the frog
A functional and structural investigation of syntactic processing in the developing brain

151 Nadine Diersch
Action prediction in the aging mind

152 Thomas Dolk
A Referential Coding Account for the Social Simon Effect

153 Mareike Bacha-Trams
Neurotransmitter receptor distribution in Broca's area and the posterior superior temporal gyrus

154 Andrea Michaela Walter
The role of goal representations in action control

155 Anne Keitel
Action perception in development: The role of experience

156 Iris Nikola Knierim
Rules don't come easy: Investigating feedback-based learning of phonotactic rules in language.

157 Jan Schreiber
Plausibility Tracking: A method to evaluate anatomical connectivity and microstructural properties along fiber pathways

158 Katja Macher
Die Beteiligung des Cerebellums am verbalen Arbeitsgedächtnis

159 Julia Erb
The neural dynamics of perceptual adaptation to degraded speech

160 Philipp Kanske
Neural bases of emotional processing in affective disorders

161 David Moreno-Dominguez
Whole-brain cortical parcellation: A hierarchical method based on dMRI tractography

162 Maria Christine van der Steen
Temporal adaptation and anticipation mechanisms in sensorimotor synchronization

163 Antje Strauß
Neural oscillatory dynamics of spoken word recognition

164 Jonas Obleser
The brain dynamics of comprehending degraded speech

165 Corinna E. Bonhage
Memory and Prediction in Sentence Processing

S 2 Tania Singer, Bethany E. Kok, Boris Bornemann, Matthias Bolz, and Christina A. Bochow
The Resource Project
Background, Design, Samples, and Measurements

166 Anna Wilsch
Neural oscillations in auditory working memory

167 Dominique Goltz
Sustained Spatial Attention in Touch: Underlying Brain Areas and Their Interaction

168 Juliane Dinse
A Model-Based Cortical Parcellation Scheme for High-Resolution 7 Tesla MRI Data

169 Gesa Schaadt
Visual, Auditory, and Visual-Auditory Speech Processing in School Children with Writing Difficulties

170 Laura Verga
Learning together or learning alone: Investigating the role of social interaction in second language word learning

171 Eva Maria Quinque
Brain, mood and cognition in hypothyroidism

172 Malte Wöstmann
Neural dynamics of selective attention to speech in noise

173 Charles-Étienne Benoit
Music-based gait rehabilitation in Parkinson's disease

174 Anja Fengler
How the Brain Attunes to Sentence Processing Relating Behavior, Structure, and Function

175 Emiliano Zaccarella
Breaking Down Complexity: The Neural Basis of the Syntactic Merge Mechanism in the Human Brain

S 2 Tania Singer, Bethany E. Kok, Boris Bornemann, Sandra Zurborg, Matthias Bolz, and Christina Bochow
The Resource Project
Background, Design, Samples, and Measurements
Second Edition

176 Manja Attig
Handlungsverständnis in den ersten Lebensjahren: retrospektive und prospektive Verarbeitung

177 Andrea Reiter
Out of control behaviors? Investigating mechanisms of behavioral control in alcohol addition, binge eating disorder, and associated risc factors

178 Anna Strotseva-Feinschmidt
The processing of complex syntax in early childhoodrs