Multimodal neuroimaging of prefrontal cortex (dys)function: EEG, fNIRS, fNIRS-fMRI and Imaging Genetics approaches

Dissertation zur Erlangung des
naturwissenschaftlichen Doktorgrades
der Julius-Maximilians-Universität Würzburg

vorgelegt von

Dipl.-Biol. Sebastian Heinzel

aus Hilden

Würzburg 2012

Eingereicht am: .....

Mitglieder der Promotionskommission:

Vorsitzender: .....

Gutachter: Prof. Dr. Andreas J. Fallgatter

Gutachter: Prof. Dr. Dr. h.c. Martin Heisenberg

Tag des Promotionskolloquiums: .....

Doktorurkunde ausgehändigt am: .....

Affidavit

(Eidesstattliche Erklärung)

According §4 Abs. 3 Ziff. 3, 5 und 8

of the „Promotionsordnung der Julius-Maximilians-Universität Würzburg"

I hereby declare that my thesis entitled "**Multimodal neuroimaging of prefrontal cortex (dys)function: EEG, fNIRS, fNIRS-fMRI and Imaging Genetics approaches**" is the result of my own work. I did not receive any help or support from commercial consultants. All sources and / or materials applied are listed and specified in the thesis.

Furthermore, I verify that this thesis has not yet been submitted as part of another examination process neither in identical nor in similar form.

Tübingen .....

Meinen Eltern in Liebe und Dankbarkeit gewidmet.

Due to publisher rights the publications of this cumulative dissertation are not included in its electronic version. Please find the abstracts and full articles in the internet using the following URLs.

Study #1:

Heinzel, S., Dresler, T., Baehne, C.G., Heine, M., Boreatti-Hummer, A., Jacob, C.P., Renner, T.J., Reif, A., Lesch, K.P., Fallgatter, A.J.*, Ehlis, A.C.*

***COMT* × *DRD4* Epistasis Impacts Prefrontal Cortex Function Underlying Response Control.**

Cerebral Cortex. 2012 May 28.

http://www.ncbi.nlm.nih.gov/pubmed/22617852

Study #2

Heinzel, S., Metzger, F.G., Ehlis, A.C., Korell, R., Alboji, A., Haeussinger, F.B., Hagen, K., Maetzler, W., Eschweiler, G.W., Berg, D., Fallgatter, A.J.

Aging-related cortical reorganization of verbal fluency processing: a functional near-infrared spectroscopy study.

Neurobiology of Aging, 2013 Feb;34(2):439-50.

http://www.ncbi.nlm.nih.gov/pubmed/22770542

Study #3

Heinzel, S.*, Haeussinger, F.B.*, Hahn, T., Ehlis, A.C., Plichta, M.M., Fallgatter, A.J.

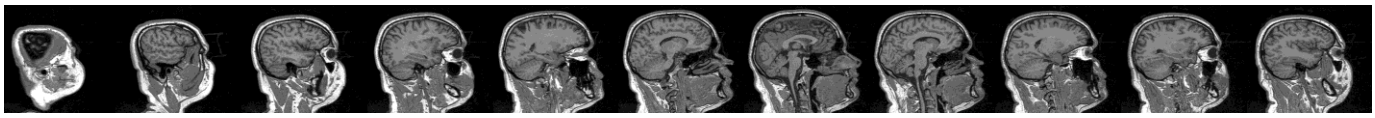
Variability of (functional) hemodynamics as measured with simultaneous fNIRS and fMRI during intertemporal choice.

Neuroimage. 2013 Jan 8;71C:125-134. doi: 10.1016/j.neuroimage.2012.12.074.

http://www.ncbi.nlm.nih.gov/pubmed/23313421

* These authors contributed equally to the study.

Content



Summary

The present cumulative dissertation comprises three neuroimaging studies using different techniques, functional tasks and experimental variables of diverse nature to investigate human prefrontal cortex (PFC) (dys)function as well as methodological aspects of functional near-infrared spectroscopy (fNIRS).

(1) Both dopamine (DA) availability ("inverted U-model") and excitatory versus inhibitory DA receptor stimulation ("dual-state theory") have been linked to PFC processing and cognitive control function. Electroencephalography (EEG) was recorded during a Go/NoGo response inhibition task in 114 healthy controls and 181 adult patients with attention-deficit/hyperactivity disorder (ADHD). As a neural measure of prefrontal cognitive response control the anteriorization of the P300 centroid in NoGo- relative to Go-trials (NoGo anteriorization, NGA) was investigated for the impact of genetic polymorphisms modulating catechol-O-methyltransferase efficiency (*COMT,* Val158Met) in degrading prefrontal DA and inhibitory DA receptor D4 sensitivity (*DRD4*, 48bp VNTR). Single genes and ADHD diagnosis showed no significant impact on the NGA or behavioral measures. However, a significant *COMT×DRD4* interaction was revealed as subjects with relatively increased D4-receptor function (*DRD4*: no 7R-alleles) displayed an "inverted U"-relationship between the NGA and increasing *COMT*-dependent DA levels, whereas subjects with decreased D4-sensitivity (7R) showed a U-relationship. This interaction was supported by 7R-allele dose-effects and also reflected by an impact on task behavior, i.e. intraindividual reaction time variability. Combining previous theories of PFC DA function, neural stability at intermediate DA levels may be accompanied by the risk of overly decreased neural flexibility if inhibitory DA receptor function is additionally decreased. The findings of *COMT×DRD4* epistasis might help to disentangle the genetic basis of dopaminergic mechanisms underlying prefrontal (dys)function.

(2) While progressive neurocognitive impairments are associated with aging and Alzheimer's disease (AD), cortical reorganization might delay difficulties in effortful word retrieval, which is one of the earliest cognitive signs of AD. Therefore, cortical hemodynamic responses were measured with fNIRS during phonological and semantic verbal fluency, and investigated in 325 non-demented, healthy subjects (age: 51-82

years). The predictive value of age, sex, verbal fluency performance and years of education for the cortical hemodynamics was assessed using multiple regression analyses. Age predicted bilaterally reduced inferior frontal junction (IFJ) and increased middle frontal and supramarginal gyri activity in both task conditions. Years of education as well as sex (IFJ activation in females > males) partly predicted opposite effects on activation compared to age, while task performance was not a significant predictor. All predictors showed small effect sizes ($-.24 < \beta < .22$). Middle frontal and supramarginal gyri activity may compensate for an aging-related decrease in IFJ recruitment during verbal fluency. The findings of aging-related (compensatory) cortical reorganization of verbal fluency processing might, in combination with other (risk) factors and using longitudinal observations, help to identify neurodegenerative processes of Alzheimer's disease, while individuals are still cognitively healthy.

(3) Individual anatomical or systemic physiological sources of variance may hamper the interpretation of fNIRS signals as neural correlates of cortical functions and their association with individual personality traits. Using simultaneous fNIRS and functional magnetic resonance imaging (fMRI) of hemodynamic responses elicited by an intertemporal choice task in 20 healthy subjects, variability in crossmodal correlations and divergence in associations of the activation with trait "sensitivity to reward" (SR) was investigated. Moreover, an impact of interindividual anatomy and scalp fMRI signal fluctuations on fNIRS signals and activation-trait associations was studied. Both methods consistently detected activation within right inferior/middle frontal gyrus, while fNIRS-fMRI correlations showed wide variability between subjects. Up to 41% of fNIRS channel activation variance was explained by gray matter volume (simulated to be) traversed by near-infrared light, and up to 20% by scalp-cortex distance. Extracranial fMRI and fNIRS time series showed significant temporal correlations at the temple. Trait SR was negatively correlated with fMRI but not fNIRS activation elicited by immediate rewards of choice within right inferior/middle frontal gyrus. Higher trait SR increased the correlation between extracranial fMRI signal fluctuations and fNIRS signals, suggesting that task-evoked systemic arousal-effects might be trait-dependent. Task-related fNIRS signals might be impacted by regionally and

individually weighted sources of anatomical and systemic physiological error variance. Trait-activation correlations might be affected or biased by systemic physiological arousal-effects, which should be accounted for in future fNIRS studies of interindividual differences.

Zusammenfassung

Die vorliegende kumulative Dissertation umfasst drei funktionelle Bildgebungsstudien, welche mit unterschiedlichen methodischen Verfahren, Versuchsaufgaben und experimentellen Variablen Hirnfunktionen des präfrontalen Kortex sowie methodische Aspekte der funktionellen Nahinfrarotspektroskopie (fNIRS) untersuchten.

(1) Sowohl die präfrontale Dopamin (DA)-Verfügbarkeit ("inverted U-model") als auch das Verhältnis der Stimulation von exzitatorischen und inhibitorischen DA-Rezeptoren ("dual-state theory") wurde mit präfrontaler Verarbeitung und Funktionen wie kognitiver Kontrolle in Verbindung gebracht. Während der Bearbeitung einer Aufgabe zur motorischen Antworthemmung wurden die elektrischen Hirnsignale mittels Elektroenzephalographie (EEG) bei 114 gesunden Probanden und 181 adulten Patienten mit Aufmerksamkeitsdefizit-/Hyperaktivitätsstörung abgeleitet. Als neuronales Maß der präfrontalen kognitiven Antwortkontrolle wurde die Anteriorisierung der P300-Zentroide während NoGo- relativ zu Go-Aufgabenbedingungen verwendet (NoGo-Anteriorisierung, NGA). Die NGA wurde hinsichtlich eines Einflusses von genetischen Polymorphismen untersucht, welche den DA Abbau durch die Katechol-O-Methyltransferase (*COMT*, Val158Met) bzw. die DA D4-Rezeptorsensitivität (*DRD4*, 48 bp VNTR) modulieren. Während die NGA weder Gen-Haupteffekte noch Unterschiede zwischen Gesunden und Patienten zeigte, war eine signifikante epistatische *COMT×DRD4* Interaktion zu beobachten. Personen mit relativ gesteigerter D4-Rezeptorsensitivität (kein 7R-Allel) zeigten einen umgekehrten U-Zusammenhang zwischen der NGA und steigender *COMT*-abhängiger DA-Verfügbarkeit, wohingegen Personen mit relativ verringerter D4-Rezeptorsensitivität (7R-Allel) einen U-Zusammenhang zeigten. Diese Gen-Gen Interaktion zeigte *DRD4* 7R-Alleldosis-Effekte und spiegelte sich auch behavioral in der intraindividuellen Go-

Reaktionszeitvariabilität wider. Neuronale Stabilität bei mittlerer DA-Verfügbarkeit könnte mit einem erhöhen Risiko verringerter Flexibilität einhergehen, wenn zusätzlich die inhibitorische DA D4-Rezeptorfunktion eingeschränkt ist. Über die gezeigte Interaktion genetischer Einflussvariablen vereinigen die Ergebnisse bestehende Theorien zur DA-Verfügbarkeit bzw. dem Verhältnis DA-abhängiger neuronaler Erregung und Hemmung mit Einfluss auf präfrontale kognitive Kontrolle.

(2) Alterungsprozesse und die Alzheimer-Demenz sind mit Beeinträchtigungen neurokognitiver Funktionen verbunden, wobei eine verringerte Wortflüssigkeit zu den frühesten Symptomen der Alzheimer-Demenz gehört. Kompensatorische Prozesse, welche diesen Symptomen (zunächst) entgegenwirken, können sich in einer Reorganisation kortikaler Verarbeitung zeigen. Zur Untersuchung dieser Prozesse wurden kortikale hämodynamische Antworten während der phonologischen und semantischen Wortflüssigkeit wurden bei 325 nicht-dementen gesunden Personen (Alter: 51-82 Jahre) mittels fNIRS untersucht. Der prädiktive Wert von Alter, Geschlecht, Wortflüssigkeitsleistung und der Ausbildungsjahre der Versuchspersonen bezüglicher der kortikalen hämodynamischen Antworten wurde mittels multipler Regression untersucht. Das Alter war ein signifikanter Prädiktor reduzierter bilateraler Aktivität im Übergangsbereich vom inferior frontalen Gyrus zum temporalen Pol (IFT) und gesteigerter bilateraler Aktivität im mittleren frontalen und supramarginalen Gyrus. Die Ausbildungsjahre und das Geschlecht (IFT-Aktivität bei Frauen höher als bei Männern) zeigten teilweise dem Alter entgegengesetzte Effekte, während die Wortflüssigkeitsleistung keinen signifikanten Einfluss hatte. Alle Prädiktoren zeigten nur kleine Effektstärken ($-.24 < \beta < .22$). Die gesteigerte Aktivität im mittleren frontalen und supramarginalen Gyrus könnte einen Kompensationsprozess für gesenkte IFT Aktivität mit steigendem Altern darstellen. Diese Belege einer (kompensatorischen) kortikalen Reorganisation der Verarbeitung von Wortflüssigkeit könnten, in Kombination mit weiteren (Risiko-)Faktoren und im Rahmen longitudinaler Untersuchungen, dazu beitragen neurodegenerative Prozesse einer Alzheimer-Demenz zu erkennen, bevor erste kognitive Symptome erkennbar sind.

(3) Einflüsse individueller Anatomie und systemischer physiologischer Artefakte können die Validität der Interpretation von fNIRS Signalen als Korrelate kortikaler Hirn-

aktivität und Korrelationen dieser Aktivität mit individuellen (Persönlichkeits-)Maßen einschränken. Zur Untersuchung dieser Problematik wurde eine simultane Messung hämodynamischer Antworten mit fNIRS und funktioneller Magnetresonanztomographie (fMRT) bei 20 gesunden Versuchspersonen durchgeführt, während eine Entscheidungsaufgabe zwischen Geldbeträgen unterschiedlicher Höhe und Aushändigungszeitpunkte durchgeführt wurde. Beide Methoden zeigten konsistente Aktivierung im rechten inferioren/mittleren frontalen Gyrus. Korrelationen der fNIRS mit den fMRT Zeitreihen zeigten jedoch eine hohe Variabilität zwischen den Versuchspersonen. Bis zu 41% der Varianz der fNIRS-Aktivität wurde durch das simulierte individuelle Volumen der von fNIRS erfassten grauen Hirnsubstanz eines Messkanals, und bis zu 20% durch den Abstand zwischen Kopfoberfläche und Kortex, aufgeklärt. Die fMRT-Zeitreihen in der Haut zeigten zudem signifikante Korrelationen mit dem fNIRS-Signal in der Schläfenregion. Während fMRT eine signifikante negative Korrelation der inferioren/mittleren frontalen Gyrus-Aktivität mit dem Persönlichkeitsmerkmal "Belohnungssensitivität" zeigte, war die Korrelation bei fNIRS nicht signifikant. Eine erhöhte Belohnungssensitivität erhöhte zudem die Korrelation zwischen fNIRS und fMRT in der Haut, welches auf eine durch Erregung erhöhte systemisch-physiologische Reaktion in Abhängigkeit des Persönlichkeitsmerkales hindeuten könnte. Die mit fNIRS aufgezeichneten hämodynamischen Antworten unterliegen regionaler und individuell-gewichteter anatomischer und systemisch-physiologischer Fehlervarianz und zukünftige fNIRS-Studien zu interindividuellen Unterschieden sollten diesen Umstand berücksichtigen.

Abbreviations

ACC	**Anterior cingulate cortex**
AD	**Alzheimer's disease**
BOLD	**Blood-oxygenation-level-dependent**
CNS	**Central nervous system**
COMT	**Catechol-O-methyltransferase**
CSF	**Cerebrospinal fluid**
DA	**Dopamine**
Deoxy	**Deoxygenated hemoglobin**
DLPFC	**Dorsolateral prefrontal cortex**
DNA	**Deoxyribonucleic acid**
DSM	**Diagnostic and Statistical Manual of Mental Disorders**
DTI	**Diffusion tensor imaging**
EEG	**Electroencephalography**
EPI	**Echo-planar imaging**
ERP	**Event-related potential**
FDR	**False discovery rate**
fMRI	**Functional magnetic resonance imaging**
fNIRS	**Functional near-infrared spectroscopy**
FWE	**Family-wise error**
GABA	**γ-Aminobutyric acid**
HHb	**Deoxygenated hemoglobin**
IFJ	**Inferior frontal junction**
ITC	**Intertemporal choice**

MCI	**Mild cognitive impairment**
MRI	**Magnetic resonance imaging**
NGA	**NoGo-anteriorization**
O_2Hb	**Oxygenated hemoglobin**
OFC	**Orbitofrontal cortex**
Oxy	**Oxygenated hemoglobin**
PFC	**Prefrontal cortex**
ROI	**Region of interest**
RT	**Reaction time**
SCD	**Scalp-cortex distance**
SCID	**Structured interview for DSM-IV**
SD	**Standard deviation**
SEM	**Standard error of the mean**
SNP	**Single nucleotide polymorphism**
SR	**Sensitivity to reward**
VBM	**Voxel-based morphometry**
VFT	**Verbal fluency test**
V_{gray}	**Volume of gray matter absorbing light**
VMPFC	**Ventromedial prefrontal cortex**
VNTR	**Variable-number of tandem repeat**

General introduction

The peripheral and central nervous system (CNS) serve a plethora of functions, which ultimately regulate the body system and its interaction with the environment. Information processing mediated by neural networks are fundamental to these functions – from basic cellular and physiological homeostasis of various organ systems, sensory processes, cognitive functions underlying complex decision-making processes, to motor processes underlying behavior. Thereby, these neural systems allow animals to adapt and respond as relevant intrinsic or external parameters of the functioning of body or mind change over time.

"… to move things is all that mankind can do, for such the sole executant is muscle, whether in whispering a syllable or in felling a forest." (Charles Sherrington, 1924)

In higher vertebrate and especially humans, the prefrontal cortex (PFC) plays a central role for the neural processes of cognitive control and (executive) functions underlying behavior (Fuster, 2008a; Miller and Cohen, 2001). Cognition and behavior are vastly defined by the neural networks of the PFC and its connectivity with other brain regions, which have been sculpted by environmental and (epi)genetic influences.

The present cumulative dissertation comprises studies of various neuroimaging techniques investigating cognitive control and executive functions via task-related activation of the PFC and its modulation by (1) molecular genetic factors and psychopathology, (2) demographic and behavioral factors, and (3) personality traits. Moreover, optical neuroimaging of PFC functional hemodynamics is examined in regard to potential confounding factors of systemic physiological hemodynamic influences and an impact of individual anatomy. These different factors investigated in the light of PFC activation and function, as well as neuroimaging methodology, are of highly diverse nature in respect to their role within the principal organization of the human CNS. To give a general overview of the conceptually discrete, yet functionally highly interconnected entities – from genes to personality – the principal organizational

levels of the human CNS are briefly introduced: From the genetic and molecular level to cells to cellular networks to brain functions, behavior and personality.
Thereafter, an introduction to the PFC and its functions as well as neuroimaging methods and approaches involved in the studies of the present cumulative dissertation is given.

Organizational levels of the CNS

While the genetic information in form of deoxyribonucleic acid (DNA) principally codes for protein elements of all cells, many possible mechanisms may regulate the expression of a gene coding for a particular protein. The emerging patterns of gene expression are specific to cell types, and thereby largely define the cellular phenotypic identity. Gene expression is modulated at the level of transcription, mRNA processing and translation, however, involving a multitude of (epi)genetic mechanisms such as the regulation of DNA-histone complexation and DNA methylation, the binding of transcription factors and other proteins to the DNA and various post-transcriptional regulations (Graw, 2006). Therefore, (epi)genetic mechanisms play a key role in determining cellular function. The (epi)genetic factors can, for instance, impact on neuronal or glial function by modulating differentiation, growth and morphogenesis, "housekeeping" function, (synaptic) plasticity, and neurotransmission (Bilder et al., 2004; Fagiolini et al., 2009; Homberg and Lesch, 2011; Olynik and Rastegar, 2012). Also, genetic variability mediated by single nucleotide polymorphisms (SNPs), variable-number-of-tandem-repeat (VNTR) polymorphisms, gene copy number variation (CNV), microsatellites, and deletions/insertions of DNA regions may either change transcriptional activity or translational efficiency, or protein conformation and function. Thereby, genetic variants can modulate various functional processes of cellular function, such as neurotransmission. In this regard, a functional role of genetic variants has been implicated for, e.g. neurotransmitter synthesis (*TPH2*), vesicular release (*SNAP25*), synaptic reuptake (*5-HTT*, *DAT*) or catabolism (*COMT, MAOA*), as well as post- and presynaptic receptor function (*DRD4*, *DRD2*),

ion channel function (*KCNJ6*) or signal transduction cascades (*DARPP32*). Moreover, genetic variation of genes for neurotrophic factors (*BDNF*), plasticity/transcription factors (*CREB1*), neuropeptides (*NPY*, *NPS*), apolipoproteins (*APOE*) and many others likely impact important processes of cellular or CNS function (Allen et al., 2008; Arcos-Burgos et al., 2012; Gizer et al., 2009; Levinson, 2006; Miyakawa, 2007).

The organizational level of molecular (epi)genetics (in interaction with the extracellular environment) defines the molecular, morphological and functional identity of cells. The cellular phenotypic details allow for a differentiation of up to 411 distinct cell types in the adult human body, with at least 145 types of neurons in the adult human brain (Vickaryous and Hall, 2006). However, each cell is unique and constantly changing. Thus, the individuality of each human brain and each individual personality may only in part be emergent from the mere number of CNS cells and the vast number of possible structural connections and functional interactions. The adult human brain has recently been estimated to contain 86 billion neurons, with 16.3 billion in the cerebral cortex, and 69 billion in the cerebellum (Herculano-Houzel, 2009). Overall, glia cells represent approximately half of all brain cells, however, large inter-regional differences in regard to the glia-to-neuron ratio have been reported (e.g., cerebral cortex gray matter: 2:1, cerebellum: 1:25, thalamus: 17:1) (Andersen et al., 1992; Azevedo et al., 2009; Pelvig et al., 2008). Estimates of an average of 7000 chemical synapses connecting onto a neocortical neuron have been reported for the (young) adult brain (Pakkenberg et al., 2003). Neurons can transfer information to other cells (or to themselves via autoreceptors or recurrent axons) using a multitude of different chemical agents, including neurotransmitters, neuropeptides, cannabinoids, and gaseous transmitters. About 100 different peptides are known to be released by different populations of neurons in the mammalian brain (See: Neuropeptide Database, an internet resource summarising all known neuropeptides, their genes, precursors and expression in the brain: http://www.neuropeptides.nl). Major neurotransmitters are acetylcholine, glutamate and γ-aminobutyric acid (GABA), and furthermore, dopamine, noradrenaline and serotonin, which often have a neuromodulatory rather than a direct neurotransmission function. In regard to their

(postsynaptic) effect neurotransmitters can be categorized as inhibitory or excitatory with glutamate being the most abundant excitatory and GABA being the major inhibitory neurotransmitter in the (adult) vertebrate CNS (Kandel et al., 2000a). However, depending on postsynaptic receptors, the same neurotransmitter may have excitatory, inhibitory as well as modulatory functions in (synaptic) neurotransmission. Specifically, the local effect of released chemical agents is defined and mediated by the transient binding to specific ionotropic or metabotropic receptor types. Most commonly, synaptic boutons release transmitters onto dendritic spines where the binding to (1) ionotropic receptors triggers the opening of ion channels and the subsequent flow of ions alters the electrochemical membrane potential. Thereby, temporal and spatial information is integrated into the dendrite. The change in membrane polarization may, in concert with other inputs, sufficiently depolarize the soma's axon hillock to generate an action potential propagating along the axon, which then itself might trigger the synaptic release of (neuro)transmitters onto postsynaptic partners. (2) Metabotropic receptors trigger intracellular (second-messenger) signaling cascades modulating, for instance, ion-channel activation/inhibition or gene transcription (Kandel et al., 2000b). The potential effects of the multitude of chemical signaling agents are multiplied by the diversity of receptor- and ion channel-isoforms (including alternative splicing variants) with specific binding, conformational and kinetic characteristics. Gene expression of different receptor types may not only be specific to particular brain regions, but stages of neural development are accompanied with the expression of, for instance, different glutamate and GABA-receptor isoforms and receptor subunit compositions (Lujan et al., 2005).

Glial cells such as astrocytes play an important role in physical support, oxygen and glucose supply via neurovascular coupling and myelin insulation of neurons. Also, accumulating evidence suggests a central role for astrocytes in the control of neuronal synaptic transmission (Haydon and Carmignoto, 2006), adding another dimension to the complexity of neural processing. Recently, astrocytes have been shown to release neurotransmitters and peptides in vicinity to neuronal synapses, e.g. in response to declining extracellular calcium levels, thereby reciprocally communicating and modulating neuron-astrocyte transmission (Araque and Navarrete; Torres et al.,

2012). Moreover, neurons as well as astrocytes often communicate through gap junctions (electrical synapses) connecting the cytoplasm of neighboring cells. This allows for an exchange of (calcium) ions or small molecules such as adenosine triphosphate (ATP). For hippocampal and neocortical neurons gap junctions, amongst other functions, are involved in the synchronization of rhythmic oscillations of activity important for memory formation and consolidation (Dere and Zlomuzica, 2012).

Sensory perception, motor behavior, every thought and mental representation and, ultimately, the individual personality of a human being is the result of information processing mediated by interconnected neuronal and glial cell assemblies and networks within the CNS and the peripheral nervous system.
150,000 to 180,000 km cumulative length of myelinated nerve fibers have been reported for the (young) adult brain (about 80,000 km at an age of 80 years) enabling extensive interneuronal information transfer (Marner et al., 2003).
The comprehensive structural description of these networks of elements and connections in the human brain is referred to as the human connectome (Sporns et al., 2005). The structural connectivity of the brain encompasses multiple scales of organization. From the microscale of cells and synapses to small neural networks to macroscopic nerve fiber bundles and morphological characteristics of interconnected brain regions (Sporns, 2011).
Despite the vast complexity of neural networks some general structural characteristics of the brain are apparent. First, for the mammalian cerebral cortex six distinctive layers can be defined based on cytoarchitecture. Each layer is composed of dendritic, somatic or axonal compartments of different neuronal cell types, such as glutamatergic projection neurons and GABAergic local interneurons. Importantly, the laminar structures differ in connectivity of projection neurons to e.g., subcortical, intracortical or thalamic regions (Kandel et al., 2000c). Regions of the cerebral cortex do not differ in the general layer composition but in the prominence of particular layers. Second, a vast literature describes a modular neocortical organization in the connection of neurons in forms of columns of about 80 to 100 neurons. However, concepts and definitions of these minicolumns as well as of macro-, and hyper-

columns vary between species, brain regions, and response properties, shared input, and common output. Therefore, the fundamentals of the function, the structural varieties and common processing mechanisms of columnar organization are far from being understood (DeFelipe et al., 2012; Horton and Adams, 2005). Recently, synaptic clustering in rats has been shown to organize a few dozen neocortical (pyramidal) neurons into mini-circuits (Perin et al., 2011). Within these 'elementary' neural building blocks the connectivity between cells had less than two degrees of separation while the number of connections was directly proportional to the number of common neighbor cells. Importantly, the connections were innate in all investigated animals rather than formed by experience, suggesting that these mini-circuits might be combined into high order constructs underlying acquired experience or memory.

On a more macroscopic level of interregional nerve fiber connections, for instance, computational tractography and diffusion tensor imaging (DTI) using non-invasive magnetic resonance imaging (MRI) have been used to generate atlases of the human connectome (Mori et al., 2009). Briefly, a set of six major network modules has been identified within the frontal, temporoparietal and medial cortex (Hagmann et al., 2008). These modules are coupled through highly connected hub-nodes largely positioned along the anterior-posterior medial axis including rostral and caudal anterior cingulate cortex, paracentral lobule and precuneus (Gong et al., 2009; Iturria-Medina et al., 2008). A similar network and hub distribution has been found for the cat and macaque monkey CNS (Zamora-Lopez et al., 2010).

While many consistencies between structural and functional connectivity of network modules and hubs have been shown (Greicius et al., 2009), it is important to note that the functional connectivity between two brain regions reflects a combination of direct and indirect network paths. Thus, structural connectivity does not rigidly determine neural dynamic interactions, but reduces the (spatial) dimensionality in which neural states can be represented, thereby allowing for fluid and variable neural processing which is sensitive to neural perturbations (Sporns, 2011). Moreover, the structural and functional neural levels are reciprocally linked through a variety of mechanisms of plasticity (Rubinov et al., 2009). Importantly, in addition to the association of clinical conditions such as Alzheimer's disease or schizophrenia to altera-

tions in hub and network structures, the connectome shows large interindividual variability. Currently, several major projects investigate the micro- and macroscopic levels of the connectome in humans and other mammals, its genetic heritability, its relation to neural processing mechanisms, dynamic patterns of resting state and task-related brain activation, characteristic alterations in neurological and psychiatric conditions and the interindividuality of connectomes, to ultimately connect the organizational levels of the brain to behavior, cognition and personality.
For recent developments in major human neuroscience and brain mapping projects see, e.g.: www.humanconnectomeproject.org
www.brain-map.org
www.humanbrainproject.eu

The psychological study of mental functions and behavior can be inherently linked to both, the science of their (developing) biological substrates as well as to, for instance, sociology, philosophy, cultural sciences, individual and cultural history, and many facets of environmental and evolutionary influences. To quantitatively relate individual cognitive processes, (personality) traits or behavior to physiological processes, various experimental psychological methods can be used. For instance, psychometric assessments can involve self-report inventories based on personality theories, clinical symptoms or behavior. Another approach is the operationalization of behavior using standardized empirical observations. Depending on the task design and the dependent and independent variables these experiments may investigate and quantify behavioral and cognitive processes. Many of these behavioral and cognitive experiments can be adapted to experimental designs suited for the simultaneous measurement of various physiological responses which precede, underlie or follow these behavioral or cognitive processes. Beside various functional neuroimaging methods assessing (surrogate) measures of neural processes in the brain, many peripheral physiological parameters can be assessed such as heart rate, blood pressure, (facial) muscle or eye movements, skin temperature and conductance, or hormone levels to name but a few.

The degrees of freedom which may underlie individual variability are countless as they emerge from the bottom of each of the organizational entities of the brain as well as through their interaction. Despite this complexity, the engagement of specific brain regions and their interaction with other regions has been identified to underlie functional roles for behavior and cognition. Herein, the PFC plays a pivotal role.

The prefrontal cortex (PFC)

The PFC comprises several widely interconnected neocortical areas with reciprocal projections from virtually all sensory systems, motor systems including (sub)thalamic and cerebellar connections, and many subcortical regions such as limbic and midbrain structures involved in reward, memory and emotional processing (Fuster, 2008b). The extensive reciprocal connectivity with other neural systems enable, for instance, multimodal convergence of visual, somatosensory, and auditory information, and direct or indirect integration of this information into the PFC, and the top-down modulatory control of the PFC over other regions, which is key to its diverse functions (Miller and Cohen, 2001; Miller et al., 2002).
While distinct PFC areas can be differentiated by morphological or cytoarchitectonic characteristics, PFC compartmentalizations may not strictly indicate their functional specialization, which rather emerges from the cooperative interaction, i.e. the structural and functional connectivity, of a particular region with other neural structures (Fuster, 2008d). Beside specific roles which have been implicated for some distinct PFC compartments (e.g., Broca area, (pre)motor area, frontal eye fields), a rough functional distinction between lateral, medial and ventral (orbitofrontal) PFC is well supported by differences in connectivity patterns and a wealth of lesion, cellular recording, neuroimaging and neuropsychological studies in primates and humans (Fuster, 2008c).

The lateral PFC plays an important role in executive behavioral control, which is enabled by (1) the collection and integration of information by its direct connections to

association cortex, limbic cortex, and subcortical structures or indirect connections through orbital and medial PFC structures, (2) the modulation and adaptive control of information flow through cortical and subcortical structures, and (3) the connections of the lateral PFC to premotor areas, the basal ganglia, and the cerebellum which support key aspects of motor behavior. Thereby, the lateral PFC encompasses a multitude of functions as for instance, attentional control, working memory, decision-making and integrative goal-directed action planning and selection, response inhibition, and emotion- and reinforcer-based behavioral control (Tanji and Hoshi, 2008). Dysfunction of the lateral PFC or its functional/structural connectivity to other regions has been implicated in psychopathological symptoms (and comorbidities of pathologies), such as self-regulation failure of appetitive behaviors in addiction, emotion regulation in depression, or deficits in various aspects of cognitive/executive functions in Alzheimer's disease, schizophrenia or ADHD (Heatherton and Wagner, 2011; Millan et al., 2012).

The orbitofrontal cortex (OFC) and parts of the ventromedial PFC (VMPFC) have dense connections with all sensory areas, limbic structures such as extensive reciprocal connections to the amygdala, insula and hippocampus and (ventromedial) striatum, moreover to the thalamus, hypothalamus, brainstem and dorsolateral PFC (Barbas, 2007; Cavada et al., 2000). Corresponding to these structural connections the OFC/VMPFC represents an important nexus for sensory integration, emotional processing, and hedonic experience (Kringelbach and Rolls, 2004), but also for reward- and punishment-guided learning, (subjective) evaluation, decision-making and maintenance of successful choices, as well as regulation of autonomic functions (Berridge and Kringelbach, 2008; Grabenhorst and Rolls, 2011; Noonan et al., 2012; Schoenbaum et al., 2011). While the OFC/VMPFC is a highly complex structure in regard to its functional interactions with other regions, its dysfunction has been linked to, for instance, impulsivity/compulsivity, addiction, obsessive–compulsive disorder (Robbins et al., 2012), and affective dysregulation, such as bipolar disorder and major depression (Cotter et al., 2005).

The (dorso)medial PFC including the anterior cingulate cortex (ACC) is integrated in both cognitive-behavioral neural networks and emotional-autonomic-motor networks

(Bush et al., 2000), and is thereby involved in emotional as well as cognitive processing. For instance, the ACC is crucial for the monitoring for processing and response conflicts and errors, and for mediating the recruitment of control functions of the DLPFC which may implement appropriate behavioral adjustments (Ridderinkhof et al., 2004a; Ridderinkhof et al., 2004b). Moreover, the (dorso)medial PFC and the ACC are important for the processing of emotional conflict regulation, social cognition and pain (Amodio and Frith, 2006; Etkin et al., 2011). For bipolar disorder and schizophrenia the medial PFC/ACC has been shown to be functionally decoupled from the DLPFC, while in bipolar disorder additionally a stronger coupling of this region and the insula and ventrolateral PFC was shown, indicating the cognitive and emotional deficits symptomatic for these disorders (Chai et al., 2011).

The studies included in the cumulative dissertation largely focused on the investigation of prefrontal cognitive control and executive functions involving the lateral PFC and the ACC. Neural correlates of these prefrontal functions were investigated in different task situations during the functional neuroimaging measurements: (1) Motor response inhibition requiring quick button response to "Go" stimuli and inhibition of that response to "NoGo" stimuli, (2) verbal fluency involving the effortful retrieval of lexical representations corresponding to phonological or semantic criteria, and (3) intertemporal choice between monetary reward options which differed in the amount and delay-to-delivery, i.e. smaller/sooner versus larger/later rewards.

Techniques for the investigation of neural functions

Over the last decades, major strides in neuroscience and its many subdisciplines were often preceded by methodological advancements regarding measurement and computation techniques as well as analytical methods and experimental designs. Various methods allow for the investigation of neural functions at different levels. By using invasive voltage/patch-clamp or intra- and extracellular electrophysiological recordings, calcium imaging or optogenetic approaches, the activity of neurons can

be directly investigated (Scanziani and Hausser, 2009). While these invasive techniques are mostly used in animal research, they allow to temporally and spatially precisely study the behavior of neuronal activity in the micro- to millisecond range. Non-invasive recordings of electrical neural activity suited for human research, such as electroencephalography (EEG) or magnetoencephalography (MEG), however, record the combined electrical activity of large neuronal assemblies comprising many thousands of neurons (Bagic and Sato, 2007). Other non-invasive modalities use surrogate measures of neural activity such as the blood oxygenation level dependent (BOLD) signal. Through astrocyte-mediated neurovascular coupling of neural activity with a local vascular response increasing blood flow, volume and oxygenation, the neural activity can be indirectly monitored (Logothetis, 2002). This process limits the temporal resolution to several seconds thereby putting a cap on the temporal resolution of methods such as functional magnetic resonance imaging (fMRI) (Bandettini, 2007) or functional near-infrared spectroscopy (fNIRS) (Ferrari and Quaresima, 2012; Hoshi, 2003; Plichta et al., 2007; Steinbrink et al., 2006). The spatial resolution in the millimeter range for fMRI and centimeter range for fNIRS only allows inferring neural function of relatively large neural assemblies comprising several million neurons. Using voxel-based morphometry (VBM) or DTI in MRI the technique also allows for structural and connectivity analyses of the brain. Moreover, positron emission tomography (PET) or single-photon emission computed tomography (SPECT) measure the isotope decay of radioactively labeled molecules such as glucose or H_2O, which can be detected in the brain after injection into the bloodstream. Thereby, metabolism or blood flow indicative of a local increase in neural activity can be studied with a spatial resolution of millimeters and a temporal resolution of several minutes (Gulyás and Sjöholm, 2007). Considering the experimental design of a functional task, stimulation or resting state performed during the functional neuroimaging recordings may allow to indirectly investigate specific brain functions, neural processing and engagement of brain regions during the experiment.

The neuroimaging techniques used in the studies of this cumulative dissertation – EEG, fNIRS and fMRI as well as Imaging Genetics – are briefly introduced.

Electroencephalography (EEG)

EEG can record the collective electrical activity resulting from extracellular ionic current flows mediated by neuronal assemblies comprising tens of thousands of (pyramidal) neurons. Voltage changes (~ 10-100 µV) recorded between EEG electrodes on the scalp surface mostly reflect action potentials propagating along myelinated axons oriented radially relative to the scalp. The electrical potential between an electrode recording neural activity and a reference electrode in an inactive area (unipolar recording), or between two electrodes recording neural activity (bipolar recording), can be measured. However, localization of the electrical source faces the "inverse problem" in EEG, i.e. the extracranially recorded potentials can have many different possible sources within the brain and the spatial location of the source of the EEG signal can only be (ambiguously) estimated following multiple assumptions about the impact of these possible generators on the recorded signal ("forward problem"). Continuous recordings of the multiple EEG channels can be analyzed with respect to their wave patterns and frequency power spectra to infer neural states from neural oscillations and spike rates in resting state or during the performance of a particular task (Bagic and Sato, 2007). Specific task-related neural functions are often investigated using event-related potentials (ERPs). Here, the temporal sequence of task events is used to average segments of EEG signals during different experimental conditions. Thereby, signals unrelated to the event are averaged out and positive and negative voltage ERP deflections in the millisecond range following the onset of a task condition can be interpreted with respect to the task characteristics to infer neural functions. Characteristic latencies of an ERP waveform peak as well as its amplitude (1-30 µV) are often related to specific neural processes (Birbaumer and Schmidt, 2006). For instance, the P300 (positive deflection at roughly 250-500 ms after the onset of an event) has been associated with attentional processes sensitive to task processing demands and individual differences in cognitive capability. Specifically, the P300 amplitude has been associated with attentional resource allocation implicating cognitive demands during task processing (Polich, 2007).

Functional magnetic resonance imaging (fMRI)

Two major fundamental principles underlie fMRI. First, after (anti)parallel alignment of the spin vector of protons to a magnetic field the vector begins to precess in the presence of a second orthogonal magnetic field. Magnetic resonance occurs when applying radio frequency (rf) pulses with the same frequency as the proton precession, thereby increasing the nuclear spin energy (magnetization), as indicated by the flip angle of the precession vector. When switching off the rf-pulses the precession vector returns to the equilibrium state within a certain amount of time (relaxation time). The reduction of magnetization induces a current in a (receiver) rf-coil, which in combination with the relaxation times indicate physical and chemical characteristics (proton density) unique to certain tissue types of the body. To make an image, the spin's precession frequency has to be made dependent on the location of the spin, which is achieved by using different magnetic field gradients. Slice selection, and phase and frequency encoding allow for an image formation of the data recorded by the rf-coil. Ultimately, about 50.000 voxels of 1 mm^3 or smaller, depending on the maximal field strength of the MRI scanner, generate a structural image of the brain, which can be used for structural analyses such as VBM (e.g. cortical gray matter thickness, hippocampal volume, etc.) or as a template to superimpose functional images (Bandettini, 2007).

Second, fMRI is based on the differences in magnetic properties between oxygenated (O_2Hb) and deoxygenated hemoglobin (HHb). HHb is paramagnetic, whereas O_2Hb has the same magnetic susceptibility as water and brain tissue. Thus, the presence of HHb creates a local distortion of the magnetic field, and since the field strength is directly proportional to the precession frequency of protons, their precession coherence is disturbed through field inhomogenities (due to HHb) causing a relative decrease in the MRI signal. During neural activation the blood flow increases locally causing a decrease in the amount of HHb, and thus, a relative MRI signal increase of a few percent (Bandettini, 2007). This increase in the BOLD-signal can be related to the temporal sequence of a functional task condition performed during the fMRI measurements, thus indirectly indicating task-related neural activation. However,

the BOLD-response is relatively slow and is usually only detected after several seconds (~ 4-7 s) from the onset of the underlying neural activity, thereby limiting the temporal resolution of fMRI (Rosen et al., 1998).

Functional near-infrared spectroscopy (fNIRS)

fNIRS is an optical neuroimaging method that exploits (1) the transparency of biological tissues for light of wavelengths between 600-1000 nm, and (2) the characteristic absorption spectra of O_2Hb and HHb, respectively, allowing for spectroscopic differentiation. (3) Importantly, changes of the physiological state of the cerebral tissue result in negligible changes in light scattering (Obrig and Villringer, 2003; Obrig et al., 2000). Using a modified Beer Lambert law, relative concentration changes can be calculated as a function of total photon path length (Villringer and Chance, 1997). Since the length of the optical path cannot be measured by continuous wave systems, the scale unit of the change in O_2Hb and HHb, respectively, equals the concentration multiplied by the unknown path length [mmol × mm] (Hoshi, 2003). Since the path length and the tissue composition traversed by the light might differ between scalp positions, signals cannot be quantitatively compared between different fNIRS channels.

Using continuous-wave fNIRS systems, near-infrared light is constantly emitted traversing the highly scattering media of scalp, scull, cerebrospinal fluid and cortical tissue. Due to scattering and absorption, only about 0.001% of the emitted light reaches the detector which is commonly positioned on the head surface 3 cm apart from the emitter (Haeussinger et al., 2011; Okada et al., 1997). The light follows an ellipsoid path through the tissue, traversing cortical gray matter in a depth of 2-3 cm from the head surface before reaching the detector (Cui et al., 2011; Haeussinger et al., 2011). The relative changes in O_2Hb and HHb concentrations in the gray matter change the light absorption, and thereby the light intensity measured at the detector. Similar to fMRI, the BOLD-signal thereby indicates task-related neural activation of the cortex. Compared to fMRI, fNIRS has a lower spatial resolution of about 2-3 cm,

and while the fNIRS sampling rate can be sampled above 10 Hz, the relatively slow BOLD-response is a temporal limitation of measuring neural events.
To record fNIRS signals, a probe-set is positioned on the scalp which consists of, for instance, 17 light emitters and 16 detectors with 3 cm inter-optode distance creating 52 recording channels (each ~3 cm spatial resolution) covering a cortical area of interest of 6 cm × 30 cm. The benefit of fNIRS (compared to fMRI) lies in its ease of use, low cost, ecological validity (no noise, comfortable measurements in a more natural environment), and relatively low susceptibility to motion artifacts. Thus, fNIRS may be particularly suited to investigate neural functions in infants and in psychiatric patients. On the other hand, fNIRS has a relatively poor spatial resolution and is restricted to cortical measurements, which can be affected by confounding influences such as individual anatomy or systemic physiological artifacts decreasing the signal to noise ratio (Cui et al., 2011; Hoshi, 2007).

Imaging Genetics

To investigate whether a particular genotype has a phenotypic effect on the level of neural processing/activation or neuroanatomy as assessed with functional neuroimaging, these measures are statistically investigated for differences between genotype groups of subjects. This approach is referred to as Imaging Genetics or Genomic Imaging (first described by: Fallgatter et al., 1999). Genetic effects show higher penetrance to the level of neural activation or anatomy compared to effects on behavioral phenotypes or psychopathology. Thereby, Imaging Genetics may provide insight into the genetic underpinnings of neural processing or pathomechanisms of psychiatric illnesses (Gottesman and Gould, 2003).

Each functional neuroimaging method has its own merits and limitations, and by using different modalities as well as functional tasks, the three studies included in the present cumulative dissertation yield different perspectives of prefrontal cognitive

control and executive functions, their neural correlates and aspects of methodological approaches in functional neuroimaging.

In Study #1 EEG and P300 ERP recordings were used to examine prefrontal cognitive response control during a response inhibition task in a sample of healthy controls and adult ADHD patients. The neural and behavior correlates of cognitive response control, which were hypothesized to differ between these groups, were furthermore investigated for an (epistatic) impact of two dopaminergic gene variants associated with altered PFC processing and ADHD, respectively.

In Study #2 cortical hemodynamic responses elicited by prefrontal cognitive/executive processing underlying verbal fluency were measured in elderly subjects using fNIRS. The impact of age, sex, years of education and task performance on the hemodynamic correlates of cortical verbal fluency processing was examined as part of a multidisciplinary longitudinal study aiming to identify risk factors of neurodegeneration in Alzheimer's disease and Parkinson's disease, respectively.

In Study #3 simultaneous fNIRS-fMRI measurements were used to (1) compare these neuroimaging methods regarding prefrontal hemodynamic responses during an intertemporal reward choice task, (2) investigate individual anatomical and systemic physiological factors impacting fNIRS measurements, and (3) to examine the correlation between trait "sensitivity to reward" and the fNIRS and fMRI activation measures, respectively, and the impact of sources of error variance on activation-trait associations in fNIRS data.

While the details of these studies were not in the scope of this general introduction, the specific background, hypotheses and methods used to investigate PFC (dys)function are given in the respective sections of the following publications.

References (General Introduction)

Allen, N.C., Bagade, S., McQueen, M.B., Ioannidis, J.P.A., Kavvoura, F.K., Khoury, M.J., Tanzi, R.E., Bertram, L., 2008. Systematic meta-analyses and field synopsis of genetic association studies in schizophrenia: the SzGene database. Nature Genetics 40, 827-834.

Amodio, D.M., Frith, C.D., 2006. Meeting of minds: the medial frontal cortex and social cognition. Nat Rev Neurosci 7, 268-277.

Andersen, B.B., Korbo, L., Pakkenberg, B., 1992. A Quantitative Study of the Human Cerebellum with Unbiased Stereological Techniques. Journal of Comparative Neurology 326, 549-560.

Araque, A., Navarrete, M., Glial cells in neuronal network function. Philos Trans R Soc Lond B Biol Sci 365, 2375-2381.

Arcos-Burgos, M., Velez, J.I., Solomon, B.D., Muenke, M., 2012. A common genetic network underlies substance use disorders and disruptive or externalizing disorders. Human Genetics 131, 917-929.

Azevedo, F.A.C., Carvalho, L.R.B., Grinberg, L.T., Farfel, J.M., Ferretti, R.E.L., Leite, R.E.P., Jacob, W., Lent, R., Herculano-Houzel, S., 2009. Equal Numbers of Neuronal and Nonneuronal Cells Make the Human Brain an Isometrically Scaled-Up Primate Brain. Journal of Comparative Neurology 513, 532-541.

Bagic, A., Sato, S., 2007. Principles of electroencephalography and magnetoencephalography. In: Hillary, F.G., DeLuca, J. (Eds.), Functional neuroimaging in clinical populations. The Guilford Press, New York, pp. 71-89.

Bandettini, P.A., 2007. Principles of functinoal magnetic resonance imaging. In: Hillary, F.G., DeLuca, J. (Eds.), Functional neuroimaging in clinical populations. The Guilford Press, New York, pp. 31-70.

Barbas, H., 2007. Flow of information for emotions through temporal and orbitofrontal pathways. Journal of Anatomy 211, 237-249.

Berridge, K.C., Kringelbach, M.L., 2008. Affective neuroscience of pleasure: reward in humans and animals. Psychopharmacology 199, 457-480.

Bilder, R.M., Volavka, J., Lachman, H.M., Grace, A.A., 2004. The catechol-O-methyltransferase polymorphism: relations to the tonic-phasic dopamine hypothesis and neuropsychiatric phenotypes. Neuropsychopharmacology 29, 1943-1961.

Birbaumer, N., Schmidt, R.F., 2006. Biologische Psychologie. Springer, Heidelberg, pp. 476-483.

Bush, G., Luu, P., Posner, M.I., 2000. Cognitive and emotional influences in anterior cingulate cortex. Trends Cogn Sci 4, 215-222.

Cavada, C., Company, T., Tejedor, J., Cruz-Rizzolo, R.J., Reinoso-Suarez, F., 2000. The anatomical connections of the macaque monkey orbitofrontal cortex. A review. Cerebral Cortex 10, 220-242.

Chai, X.J., Whitfield-Gabrieli, S., Shinn, A.K., Gabrieli, J.D., Nieto Castanon, A., McCarthy, J.M., Cohen, B.M., Ongur, D., 2011. Abnormal medial prefrontal cortex resting-state connectivity in bipolar disorder and schizophrenia. Neuropsychopharmacology 36, 2009-2017.

Cotter, D., Hudson, L., Landau, S., 2005. Evidence for orbitofrontal pathology in bipolar disorder and major depression, but not in schizophrenia. Bipolar Disord 7, 358-369.
Cui, X., Bray, S., Bryant, D.M., Glover, G.H., Reiss, A.L., 2011. A quantitative comparison of NIRS and fMRI across multiple cognitive tasks. Neuroimage 54, 2808-2821.
DeFelipe, J., Markram, H., Rockland, K.S., 2012. The neocortical column. Frontiers in Neuroanatomy 6.
Dere, E., Zlomuzica, A., 2012. The role of gap junctions in the brain in health and disease. Neuroscience and Biobehavioral Reviews 36, 206-217.
Etkin, A., Egner, T., Kalisch, R., 2011. Emotional processing in anterior cingulate and medial prefrontal cortex. Trends Cogn Sci 15, 85-93.
Fagiolini, M., Jensen, C.L., Champagne, F.A., 2009. Epigenetic influences on brain development and plasticity. Curr Opin Neurobiol 19, 207-212.
Fallgatter, A.J., Jatzke, S., Bartsch, A.J., Hamelbeck, B., Lesch, K.P., 1999. Serotonin transporter promoter polymorphism influences topography of inhibitory motor control. Int J Neuropsychopharmacol 2, 115-120.
Ferrari, M., Quaresima, V., 2012. A brief review on the history of human functional near-infrared spectroscopy (fNIRS) development and fields of application. Neuroimage.
Fuster, J., 2008a. The Prefrontal Cortex. Academic Press, Boston, pp. 346-355.
Fuster, J., 2008b. The Prefrontal Cortex. Academic Press, Boston, pp. 27-44.
Fuster, J., 2008c. The Prefrontal Cortex. Academic Press, Boston, pp. 27-62.
Fuster, J., 2008d. The Prefrontal Cortex. Academic Press, Boston, pp. 1-6.
Gizer, I.R., Ficks, C., Waldman, I.D., 2009. Candidate gene studies of ADHD: a meta-analytic review. Hum Genet 126, 51-90.
Gong, G.L., He, Y., Concha, L., Lebel, C., Gross, D.W., Evans, A.C., Beaulieu, C., 2009. Mapping Anatomical Connectivity Patterns of Human Cerebral Cortex Using In Vivo Diffusion Tensor Imaging Tractography. Cerebral Cortex 19, 524-536.
Gottesman, I.I., Gould, T.D., 2003. The endophenotype concept in psychiatry: Etymology and strategic intentions. American Journal of Psychiatry 160, 636-645.
Grabenhorst, F., Rolls, E.T., 2011. Value, pleasure and choice in the ventral prefrontal cortex. Trends in Cognitive Sciences 15, 56-67.
Graw, J., 2006. Genetik. Springer, Berlin Heidelberg, pp. 81-82,272-289,320-328,510-515.
Greicius, M.D., Supekar, K., Menon, V., Dougherty, R.F., 2009. Resting-state functional connectivity reflects structural connectivity in the default mode network. Cereb Cortex 19, 72-78.
Gulyás, B., Sjöholm, N., 2007. Principles of positron emission tomography. In: Hillary, F.G., DeLuca, J. (Eds.), Functional neuroimaging in clinical populations. The Guilford Press, New York, pp. 3-30.
Haeussinger, F.B., Heinzel, S., Hahn, T., Schecklmann, M., Ehlis, A.C., Fallgatter, A.J., 2011. Simulation of near-infrared light absorption considering individual head and prefrontal cortex anatomy: implications for optical neuroimaging. PLoS One 6, e26377.

Hagmann, P., Cammoun, L., Gigandet, X., Meuli, R., Honey, C.J., Wedeen, V., Sporns, O., 2008. Mapping the structural core of human cerebral cortex. Plos Biology 6, 1479-1493.
Haydon, P.G., Carmignoto, G., 2006. Astrocyte control of synaptic transmission and neurovascular coupling. Physiol Rev 86, 1009-1031.
Heatherton, T.F., Wagner, D.D., 2011. Cognitive neuroscience of self-regulation failure. Trends Cogn Sci 15, 132-139.
Herculano-Houzel, S., 2009. The human brain in numbers: a linearly scaled-up primate brain. Front Hum Neurosci 3, 31.
Homberg, J.R., Lesch, K.P., 2011. Looking on the bright side of serotonin transporter gene variation. Biological psychiatry 69, 513-519.
Horton, J.C., Adams, D.L., 2005. The cortical column: a structure without a function. Philosophical Transactions of the Royal Society B-Biological Sciences 360, 837-862.
Hoshi, Y., 2003. Functional near-infrared optical imaging: utility and limitations in human brain mapping. Psychophysiology 40, 511-520.
Hoshi, Y., 2007. Functional near-infrared spectroscopy: current status and future prospects. J Biomed Opt 12, 062106.
Iturria-Medina, Y., Sotero, R.C., Canales-Rodriguez, E.J., Aleman-Gomez, Y., Melie-Garcia, L., 2008. Studying the human brain anatomical network via diffusion-weighted MRI and Graph Theory. Neuroimage 40, 1064-1076.
Kandel, E., Schwartz, J.H., Jessell, T.M., 2000a. Principles of neural science. McGraw-Hill, New York, pp. 280-285.
Kandel, E., Schwartz, J.H., Jessell, T.M., 2000b. Principles of neural science. McGraw-Hill, New York, pp. 180-184, 222-252
Kandel, E., Schwartz, J.H., Jessell, T.M., 2000c. Principles of neural science. McGraw-Hill, New York, pp. 325-331
Kringelbach, M.L., Rolls, E.T., 2004. The functional neuroanatomy of the human orbitofrontal cortex: evidence from neuroimaging and neuropsychology. Progress in Neurobiology 72, 341-372.
Levinson, D.F., 2006. The genetics of depression: a review. Biological psychiatry 60, 84-92.
Logothetis, N.K., 2002. The neural basis of the blood-oxygen-level-dependent functional magnetic resonance imaging signal. Philos Trans R Soc Lond B Biol Sci 357, 1003-1037.
Lujan, R., Shigemoto, R., Lopez-Bendito, G., 2005. Glutamate and GABA receptor signalling in the developing brain. Neuroscience 130, 567-580.
Marner, L., Nyengaard, J.R., Tang, Y., Pakkenberg, B., 2003. Marked loss of myelinated nerve fibers in the human brain with age. Journal of Comparative Neurology 462, 144-152.
Millan, M.J., Agid, Y., Brune, M., Bullmore, E.T., Carter, C.S., Clayton, N.S., Connor, R., Davis, S., Deakin, B., DeRubeis, R.J., Dubois, B., Geyer, M.A., Goodwin, G.M., Gorwood, P., Jay, T.M., Joels, M., Mansuy, I.M., Meyer-Lindenberg, A., Murphy, D., Rolls, E., Saletu, B., Spedding, M., Sweeney, J., Whittington, M., Young, L.J., 2012. Cognitive dysfunction in psychiatric disorders: characteristics, causes and the quest for improved therapy. Nat Rev Drug Discov 11, 141-168.

Miller, E.K., Cohen, J.D., 2001. An integrative theory of prefrontal cortex function. Annu Rev Neurosci 24, 167-202.
Miller, E.K., Freedman, D.J., Wallis, J.D., 2002. The prefrontal cortex: categories, concepts and cognition. Philos Trans R Soc Lond B Biol Sci 357, 1123-1136.
Miyakawa, T., 2007. Investigating signal transduction and genes-to-behaviors pathways in psychiatric diseases: An approach using a comprehensive behavioral test battery on genetically engineered mice. Psychiatry and Clinical Neurosciences 61, S23-S23.
Mori, S., Oishi, K., Faria, A.V., 2009. White matter atlases based on diffusion tensor imaging. Current Opinion in Neurology 22, 362-369.
Noonan, M.P., Kolling, N., Walton, M.E., Rushworth, M.F.S., 2012. Re-evaluating the role of the orbitofrontal cortex in reward and reinforcement. European Journal of Neuroscience 35, 997-1010.
Obrig, H., Villringer, A., 2003. Beyond the visible--imaging the human brain with light. J Cereb Blood Flow Metab 23, 1-18.
Obrig, H., Wenzel, R., Kohl, M., Horst, S., Wobst, P., Steinbrink, J., Thomas, F., Villringer, A., 2000. Near-infrared spectroscopy: does it function in functional activation studies of the adult brain? International Journal of Psychophysiology 35, 125-142.
Okada, E., Firbank, M., Schweiger, M., Arridge, S.R., Cope, M., Delpy, D.T., 1997. Theoretical and experimental investigation of near-infrared light propagation in a model of the adult head. Applied Optics 36, 21-31.
Olynik, B.M., Rastegar, M., 2012. The genetic and epigenetic journey of embryonic stem cells into mature neural cells. Front Genet 3, 81.
Pakkenberg, B., Pelvig, D., Marner, L., Bundgaard, M.J., Gundersen, H.J.G., Nyengaard, J.R., Regeur, L., 2003. Aging and the human neocortex. Experimental Gerontology 38, 95-99.
Pelvig, D.P., Pakkenberg, H., Stark, A.K., Pakkenberg, B., 2008. Neocortical glial cell numbers in human brains. Neurobiology of Aging 29, 1754-1762.
Perin, R., Berger, T.K., Markram, H., 2011. A synaptic organizing principle for cortical neuronal groups. Proc Natl Acad Sci U S A 108, 5419-5424.
Plichta, M.M., Heinzel, S., Ehlis, A.C., Pauli, P., Fallgatter, A.J., 2007. Model-based analysis of rapid event-related functional near-infrared spectroscopy (NIRS) data: a parametric validation study. Neuroimage 35, 625-634.
Polich, J., 2007. Updating p300: An integrative theory of P3a and P3b. Clinical Neurophysiology 118, 2128-2148.
Ridderinkhof, K.R., Ullsperger, M., Crone, E.A., Nieuwenhuis, S., 2004a. The role of the medial frontal cortex in cognitive control. Science 306, 443-447.
Ridderinkhof, K.R., van den Wildenberg, W.P., Segalowitz, S.J., Carter, C.S., 2004b. Neurocognitive mechanisms of cognitive control: the role of prefrontal cortex in action selection, response inhibition, performance monitoring, and reward-based learning. Brain Cogn 56, 129-140.
Robbins, T.W., Gillan, C.M., Smith, D.G., de Wit, S., Ersche, K.D., 2012. Neurocognitive endophenotypes of impulsivity and compulsivity: towards dimensional psychiatry. Trends in Cognitive Sciences 16, 81-91.

Rosen, B.R., Buckner, R.L., Dale, A.M., 1998. Event-related functional MRI: Past, present, and future. Proceedings of the National Academy of Sciences of the United States of America 95, 773-780.
Rubinov, M., Sporns, O., van Leeuwen, C., Breakspear, M., 2009. Symbiotic relationship between brain structure and dynamics. Bmc Neuroscience 10.
Scanziani, M., Hausser, M., 2009. Electrophysiology in the age of light. Nature 461, 930-939.
Schoenbaum, G., Takahashi, Y., Liu, T.L., McDannald, M.A., 2011. Does the orbitofrontal cortex signal value? Critical Contributions of the Orbitofrontal Cortex to Behavior 1239, 87-99.
Sporns, O., 2011. The human connectome: a complex network. Year in Cognitive Neuroscience 1224, 109-125.
Sporns, O., Tononi, G., Kotter, R., 2005. The human connectome: A structural description of the human brain. Plos Computational Biology 1, 245-251.
Steinbrink, J., Villringer, A., Kempf, F., Haux, D., Boden, S., Obrig, H., 2006. Illuminating the BOLD signal: combined fMRI-fNIRS studies. Magn Reson Imaging 24, 495-505.
Tanji, J., Hoshi, E., 2008. Role of the lateral prefrontal cortex in executive behavioral control. Physiol Rev 88, 37-57.
Torres, A., Wang, F.S., Xu, Q.W., Fujita, T., Dobrowolski, R., Willecke, K., Takano, T., Nedergaard, M., 2012. Extracellular Ca2+ Acts as a Mediator of Communication from Neurons to Glia. Science Signaling 5.
Vickaryous, M.K., Hall, B.K., 2006. Human cell type diversity, evolution, development, and classification with special reference to cells derived from the neural crest. Biol Rev Camb Philos Soc 81, 425-455.
Villringer, A., Chance, B., 1997. Non-invasive optical spectroscopy and imaging of human brain function. Trends in Neurosciences 20, 435-442.
Zamora-Lopez, G., Zhou, C., Kurths, J., 2010. Cortical hubs form a module for multisensory integration on top of the hierarchy of cortical networks. Front Neuroinform 4, 1.

Cumulative dissertation of Dipl.-Biol. Sebastian Heinzel:
Declaration of own and co-author contributions

Manuscript:

"*COMT* x *DRD4* epistasis impacts prefrontal cortex function underlying response control"

Sebastian Heinzel, MSc[1,2,3,†], Thomas Dresler, PhD[1,2], Christina G. Baehne, MSc[1], Monika Heine, MD[1], Andrea Boreatti-Hümmer, MD[1], Christian P. Jacob, MD[1], Tobias J. Renner, MD[4], Andreas Reif, MD[1], Klaus-Peter Lesch, MD[1], Andreas J. Fallgatter, MD[2,*], Ann-Christine Ehlis, PhD[2,*]

[1]Department of Psychiatry, Psychosomatics and Psychotherapy, University of Wuerzburg, Germany

[2]Department of Psychiatry and Psychotherapy, University of Tuebingen, Germany

[3]German Center for Neurodegenerative Diseases (DZNE), Tuebingen, Germany

[4]Department of Child and Adolescent Psychiatry, Psychosomatics and Psychotherapy, University of Wuerzburg, Germany

*These authors contributed equally to the study.

†Corresponding author

Author contributions:

Study planning: AJF, ACE

Recruitment of participants and clinical diagnosis: CGB, MH, ABH, CPJ, KPL

EEG data acquisition: CGB

Genotyping: TJR, AR, KPL

Data analysis and interpretation: SH, ACE, CGB, TD

Writing of manuscript: SH, TD, ACE

Hereby, we confirm the author contributions listed above:

Sebastian Heinzel, MSc .....

Thomas Dresler, PhD .....

Christina G. Baehne, MSc .....

Monika Heine, MD .....

Andrea Boreatti-Hümmer, MD .....

Christian P. Jacob, MD .....

Tobias J. Renner, MD .....

Andreas Reif, MD .....

Klaus-Peter Lesch, MD .....

Andreas J. Fallgatter, MD .....

Ann-Christine Ehlis, PhD .....

Cumulative dissertation of Dipl.-Biol. Sebastian Heinzel:

Declaration of own and co-author contributions

Manuscript:

"Aging-related cortical reorganization of verbal fluency processing: a functional near-infrared study"

Sebastian Heinzel, MSc[1,2,†], Florian G. Metzger, MD[1], Ann-Christine Ehlis, PhD[1], Robert Korell[1], Ahmed Alboji[1], Florian B. Haeussinger, MSc[1], Katja Hagen, MSc[1,2], Walter Maetzler, MD[2,3], Gerhard W. Eschweiler, MD[1], Daniela Berg, MD[2,3], Andreas J. Fallgatter, MD[1], and the TREND study consortium

[1]Department of Psychiatry and Psychotherapy, University of Tuebingen, Tuebingen, Germany

[2]German Center for Neurodegenerative Diseases (DZNE), Tuebingen, Germany

[3]Department of Neurodegeneration, Hertie-Institute of Clinical Brain Research, University of Tuebingen, Tuebingen, Germany

[†]Corresponding author

Author contributions:

Planning of the TREND study in general: DB, WM, GWE, AJF,

General organization and participant recruitment of the TREND study: DB, AJF, FGM, WM, GWE

Clinical supervision of the TREND study: DB, AJF, FGM, WM, GWE

Planning of fNIRS measurements: SH, AJF, KH, FM, ACE

fNIRS data acquisition: SH, RK, AA

Database of behavioral and demographic information: SH, RK, AA

Programming of analysis tools: SH, FBH

fNIRS and behavioral/demographic data analysis and interpretation: SH

Writing of manuscript: SH

Critical revision of the manuscript by all coauthors.

Hereby, we confirm the author contributions listed above:

Sebastian Heinzel, MSc .....

Florian G. Metzger, MD .....

Ann-Christine Ehlis, PhD .....

Robert Korell, Cand. Med. .....

Ahmed Alboji, Cand. Med. .....

Florian B. Haeussinger, MSc .....

Katja Hagen, MSc .....

Walter Maetzler, MD .....

Gerhard W. Eschweiler, MD .....

Daniela Berg, MD .....

Andreas J. Fallgatter, MD .....

Cumulative dissertation of Dipl.-Biol. Sebastian Heinzel:

Declaration of own and co-author contributions

Manuscript:

"Variability of (functional) hemodynamics as measured with simultaneous fNIRS and fMRI during intertemporal choice"

Sebastian Heinzel, MSc[1,2,3,*,†], Florian B. Haeussinger, MSc[1,*], Tim Hahn, PhD[4], Ann-Christine Ehlis, PhD[1], Andreas J. Fallgatter, MD[1]

[1]Department of Psychiatry and Psychotherapy, University of Tuebingen, Germany

[2]German Center for Neurodegenerative Diseases (DZNE), Tuebingen, Germany

[3]Department of Psychiatry, Psychosomatics and Psychotherapy,
University of Wuerzburg, Germany

[4]Department of Cognitive Psychology II, University of Frankfurt am Main, Frankfurt am Main, Germany

*These authors contributed equally to the study.

†Corresponding author

Author contributions:

Planning of the study: SH, FBH, AJF

Organization and participant recruitment: SH, AJF

fNIRS and fMRI data acquisition: SH, FBH

Programming of analysis tools: SH, FBH

Data analysis and interpretation: SH, FBH, TH, ACE

Writing of manuscript: SH, FBH, TH, ACE

Critical revision of the manuscript by all coauthors.

Hereby, we confirm the author contributions listed above:

Sebastian Heinzel, MSc ….....

Florian B. Haeussinger, MSc ….....

Tim Hahn, PhD ….....

Ann-Christine Ehlis, PhD ….....

Andreas J. Fallgatter, MD ….....

Note, that Dr. Michael M. Plichta was not listed as an author at the time of the submission of the manuscript and the dissertation. He was involved in the experimental design and programming.

General discussion

The present cumulative dissertation comprises three human neuroimaging studies investigating hemodynamic or electrical correlates of prefrontal cortex (PFC) function as well as methodological aspects of functional near-infrared spectroscopy (fNIRS). Using different neuroimaging techniques, functional paradigms and diverse individual factors of molecular genetics, (neuro)anatomy and morphology, demographic factors (e.g. age, sex, years of education), behavior, personality and psychopathology, specific scientific hypotheses regarding prefrontal task-specific activation were addressed. The findings, implications and future prospects of each study are critically discussed in a general context and in the context of recent evidence and theories.

Study #1:

"*COMT*×*DRD4* Epistasis Impacts Prefrontal Cortex Function Underlying Response Control"

The study investigated the impact of two dopaminergic gene variations and their interaction on neural and behavioral correlates of prefrontal cognitive responses control using EEG and a Go-NoGo task in a large sample of 114 healthy controls and 181 adult patients with ADHD. The study showed that an epistastic interaction between *COMT*-dependent prefrontal dopamine levels and *DRD4*-dependent inhibitory D4 receptor function (*DRD4*) impacts on neural and behavioral measures of prefrontal cognitive response control.

The key hypothesis underlying this epistatic interaction is that both, (*COMT*-dependent) dopamine levels as well as differential dopamine receptor stimulation (excitatory D1 versus inhibitory D2/D4 stimulation ratio), may critically modulate prefrontal processing through an imbalance in dopamine-mediated excitation and inhibition within glutamatergic and GABAergic neural networks. Here, more stable (D1-dominated state) or more flexible (D2/D4-dominated state) neural states (Durstewitz and Seamans, 2008) may be optimal for neural and behavioral outcomes depending on the nature of a given task situation (Cools and D'Esposito, 2011). Here, the present findings suggest that subjects with intermediate dopamine-levels (associated

with stable, D1-dominated processing) have an increased risk of impaired flexibility when D4-receptor function is additionally decreased. These subjects showed reduced neural flexibility such as less efficient transitions from neural Go to NoGo representations (decreased NoGo-anteriorization) limiting performance as indicated by increased "Go" response reaction times and its intraindividual variability. Response inhibition centrally involves the anterior cingulate cortex as well as inferior frontal gyrus and cortico-thalamic motor circuits (Aron and Poldrack, 2006; Fallgatter et al., 2002). Here, exploratory source localization analyses indicated that the behavioral consequences of the epistatic effect might emerge from the level of the right premotor and supplementary motor area. However, the precise cellular effects and neural (network) dynamics which may be affected by *COMT*×*DRD4* epistasis were not investigated in this Imaging Genetics study. Specifically, the (genetic) dopaminergic modulation of the neural interaction between prefrontal excitatory glutamatergic pyramidal neurons and GABAergic interneurons was not investigated. Since ADHD patients did not significantly differ from healthy controls in regard to *COMT*×*DRD4* epistasis, the precise mechanism of this dopaminergic (gene) interaction might not be centrally involved in the pathophysiology of ADHD. However, ADHD patients showed lower P300 amplitudes at Cz and Pz positions, where the neural dynamic flexibility of P300 amplitudes in the transition from Go to NoGo trials was less pronounced compared to healthy controls. These findings may represent an endophenotype of the pathophysiology underlying deficits in response inhibition in ADHD, and it will be important to investigate the genetic and cellular basis of the altered neural processing in future studies.

In the following, further implications and possible frameworks and hypotheses for future studies derived from the present findings are given.

In addition to ADHD, dopaminergic dysregulation and deficits in prefrontal executive functions are also common for other clinical conditions such as schizophrenia (Barch and Ceaser, 2012), addiction (Koob and Volkow, 2010) or Parkinson's disease (Ko et al., 2012). Tolcapone, a pharmacological centrally and peripherally acting COMT inhibitor, has been reported to improve motor functions in Parkinson's disease

(Bonifacio et al., 2007) and might improve cognitive and negative symptoms of schizophrenia and impulsivity in addiction. Currently, several clinical trials investigate the effects of tolcapone treatment on these symptoms (search "tolcapone" at: www.ClinicalTrials.gov). Importantly, these studies specifically investigated the effect of the individual *COMT* Val158Met genotype on treatment outcome for the purpose of a personalized, genotype-informed medication in the future.

In healthy subjects tolcapone has repeatedly been shown to enhance working memory performance and increase DLPFC working memory brain action in dependence of *COMT* Val158Met genotype (Apud et al., 2007; Farrell et al., 2012). COMT inhibition improved performance and increased DLPFC activation in subjects with relatively low dopamine levels and COMT enzymatic function degrading dopamine (Val/Val genotype), whereas the opposite pharmacological effect was observed in COMT Met/Met carriers which, without medication, have relatively increased dopamine levels and activation, and better working memory performance (Apud et al., 2007; Farrell et al., 2012). Thus, patients and healthy subjects carrying the Val-alleles might benefit from pharmacological COMT inhibition while the opposite might be the case for Met/Met carriers.

Working memory centrally involves a neural stable online representation. The findings of the present study suggest that for tasks involving such stable neural processing Val-carriers might benefit from pharmacological COMT inhibition, however, subjects with additionally decreased inhibitory D4-receptor function might exhibit overly decreased neural and behavioral flexibility. Thus, in addition to *COMT* Val158Met, genotypes affecting dopamine receptor function should be considered to optimize pharmacological effects in respect to (executive) functions important for well-being and daily living. Conversely, antipsychotic treatment in schizophrenia and bipolar disorders using (non-selective) dopamine receptor (D2 type) antagonists should be further investigated for an impact of COMT genotype. A moderating role of *COMT* genotype for the effect of antipsychotic medication on different executive functions in patients with schizophrenia have been reported showing better symptom improvement for Met-allele carriers compared to Val-carriers (Bertolino et al., 2004; Woodward et al., 2007).

In addition to *COMT* and *DRD4*, many other genetic factors may sculpt the dynamics of PFC processing, the regulation of neural excitation and inhibition and ultimately behavioral and clinical phenotypes. For future Imaging Genetics and genetic association studies the present findings may provide important hypotheses. First, the study emphasizes that phenotypic differences in PFC processing dynamics and behavior might emerge through genetic epistasis while main effects of single genes may not be detected. Second, *COMT* Val158Met is possibly the most studied SNP in (psychiatric) neuroscience. However, especially in tasks or traits for which stable versus flexible processing or behavior is required, inconsistencies in findings or additional variance especially in Val/Met carriers could be explained by genetic factors modulating dopamine receptor function, such as the *DRD4* 48-bp VNTR polymorphism. Moreover, studies stratifying for *COMT* homozygotes miss genetic and functional variability in heterozygotes which might be due to interactions with *DRD4* genotype. Since heterozygotes account for about 50% of the Caucasian population, the generalizability of the impact of *COMT* genotype on neural and behavioral phenotypes might be limited in these studies. Third, biological pathways and mechanisms are highly important to select genetic variants and generate hypotheses of main and interaction effects on different phenotypic markers within the brain. Dopamine levels as well as the ratio of excitatory versus inhibitory dopamine receptor stimulation modulating glutamatergic and GABAergic neurons and stable and flexible neural states may provide hypotheses for genetic interactions such as *COMT* × *DRD4.* Imaging Genetics and genome wide association studies (GWAS) might benefit from such *a priori* hypotheses because the access to individual genetic information has grown immensely. Nowadays, some studies investigate over 500,000 SNPs for each subject and, thus, face severe multiple testing problems, especially when epistatic interactions and numerous dependent variables, e.g. 50,000 voxels in fMRI, are included in the statistical analyses (Stein et al., 2010). Complementing such rather exploratory studies, the interpretation and biological or pathophysiological relevance of significant findings might sometimes be more conclusive when following biologically and clinically informed hypotheses derived from previous findings.

Study #2

"Aging-related cortical reorganization of verbal fluency processing:
a functional near-infrared spectroscopy study"

Increased age is the greatest risk factor for neurodegenerative diseases, such as Alzheimer's disease (AD), and deficits in verbal fluency represent one of the earliest cognitive symptoms of AD. Moreover, pathological alterations on the molecular, cellular, neural network and vascular level precede the first cognitive symptoms by years or even decades (Jack et al., 2010).

Therefore, the predictive value of age as well as sex, verbal fluency performance and years of formal education of cortical functional hemodynamic responses elicited by phonological and semantic verbal fluency were investigated in the present fNIRS study. In total, data of 325 elderly, non-demented subjects between 51 and 82 years of age was analyzed as part of the fNIRS baseline assessment of the longitudinal TREND study. Aim of this study is to identify risk factors of Alzheimer's disease and Parkinson's disease, respectively, for an early detection of these neurodegenerative diseases in subjects who are still symptomatically healthy.

The investigated predictors of the cortical activation elicited by the task showed small effect sizes ($-0.24 < \beta < 0.22$) while the multiple regression models comprising all predictors explained up to 10% of the variance of fNIRS data. Age significantly predicted both, decreased bilateral inferior frontal junction (IFJ) responses and increased bilateral middle frontal gyri and supramarginal gyri responses. This reorganization of activation with increasing age might indicate an aging-related neural compensation strategy. This finding is discussed in regard to effects of the other investigated predictors and the potential of the present findings to aid an early prediction of mild cognitive impairment and AD.

Increased activation in middle frontal gyri and supramarginal gyri could indicate increased involvement of cognitive control functions (Bush and Shin, 2006; Niendam et al., 2012). Executive functions important for verbal fluency performance, such as flexibility, inhibition, initiation, and working memory as well as attention processes have been shown to activate fronto-cingulo-parietal networks comprising the pre-

frontal and parietal regions found to be more strongly activated with increasing age in the present study.

The effortful mental search for lexical representations and verbal fluency performance with increasing age might be ameliorated by increased neural recruitment subserving these executive functions, and thereby compensate potential aging or first neurodegeneration-related deficits on multiple organization levels of the brain. Some indication of such neural decline might be represented by the decrease in bilateral IFJ (functional) hemodynamic responses with increasing age. Supporting this interpretation, glucose hypometabolism assessed using positron-emission tomography in early dementia patients correlates with executive deficits of semantic fluency ("Name items in a supermarket!") in the left IFJ (Schroeter et al., 2012). However, for aging-related bilateral functional hemodynamics in the IFJ/temple area measured using fNIRS cautious interpretations of the hemodynamics as neural activation are advised; see below for critical discussion of methodological aspects of fNIRS.

Neural and behavioral processes involved in verbal fluency and cognitive/attention network activity might also be modulated by other factors partly confounded with aging. For instance, (1) depressive symptoms, (2) alertness/fatigue, (3) other motivational and psychological aspects or (4) articulatory movements during speech production (Rusz et al., 2011) and psychomotor speed (Hipp et al., 2009), e.g. in subjects with early Parkinson's disease. In part, these additional factors may be correlated with age while not being directly associated with neurodegenerative processes involved in AD.

(1) Depression has repeatedly been associated with bilateral prefrontal hypoactivation during verbal fluency without concomitant performance deficits (Klumpp and Deldin, 2010). Depression also represents a risk factor for Alzheimer’s disease (Ownby et al., 2006), however, variance in fNIRS data due to depressive symptoms was not in the scope of the present fNIRS study. The Alzheimer’s disease risk factor of depression is investigated in the TREND study and will be examined in further fNIRS analyses. (2) In the TREND study subjects participate in eight different twenty-five minute experiments including neuropsychological testing (CERAD-Plus battery) or motor and neurological tests, which were completed in random order

within approximately three and a half hours. For some older participants the successive experiments might have been more challenging, and fatigue might have decreased performance or increased (neural) efforts to sustain attention and executive functions. Interestingly, analyses on this issue showed that the position of fNIRS within the order of the eight experiments, showed a small positive correlation ($r \sim .1$) with the phonological and semantic verbal fluency performance. While no longer significant, these correlations had a positive sign in groups of individuals over and under an age of 65 years, respectively. Therefore, participants tended to perform better after participation in other experiments, possibly because the experimental testing situation became more familiar. (3) Older subjects or subjects with a family history of AD may have been especially motivated to perform well in the verbal fluency task to disprove a personal risk of cognitive impairment. In turn, resignation or fatalistic attitudes might produce opposite effects. Thereby, increased/decreased activation of cognitive/attention fronto-cingulo-parietal networks might be psychologically explained. (4) Early Parkinson's disease can be accompanied with symptoms of altered vocal phonation and articulation (Rusz et al., 2011) as well as psychomotor speed (Hipp et al., 2009) which might impair verbal fluency or require increased neural resources underlying executive functions and attention. While these motor-related deficits differ from declining cognitive functions of effortful word retrieval, both might be correlated with increasing age.

Thus, multiple other psychological and pathological factors might be correlated with age and might be confounded with aging processes and their neural and behavioral correlates of verbal fluency. In addition to age, other variables significantly predicted (with small effect sizes) the cortical activation measured using fNIRS. Cognitive performance correlates the level of education for all ages, which may in part underlie the increased risk factor of dementia in individuals with low educational attainment levels (Caamano-Isorna et al., 2006). Consistent with these findings, years of education positively correlated with phonological ($r = .39$, $p < .001$) and semantic ($r = .15$, $p < .01$) verbal fluency performance, whereas age was only weakly correlated with semantic ($r = -.13$, $p < .05$), and did not correlate phonological verbal fluency performance.

Moreover, while no correlation between age and years of education was found, the two predictors showed partly opposing effects on the neural level. Longer education was a predictor for higher phonological verbal fluency activation in bilateral IFJ and decreased left middle frontal gyrus activation. No significant link between the cortical activation and verbal fluency performance was found suggesting that the neural effects of education might not exclusively be due to better performance. The neural differences might indicate that, for instance, (1) different cognitive strategies during effortful search for words matching the task condition criteria were employed in more educated individuals, e.g. more flexible switching between categories of syntactically connected words than clustering of words in one category, thereby generating more words (Troyer et al., 1997). (2) In addition to different behavioral strategies, more educated subjects with possibly increased crystallized and fluid intelligence might have a higher cognitive reserve, i.e. neural efficiency, capacity or compensatory recruitment of additional brain regions (Stern, 2006; Tucker and Stern, 2011). These neural mechanisms might underlie the present differences of the impact of age and education on the regional neural activation.

Also, higher education reflecting cognitive capability might reduce the risk for MCI and AD or delay their symptomatic effects (Hall et al., 2007; Sharp and Gatz, 2011), but a steeper decline in cognitive functions has been shown for more educated subjects when first AD symptoms occur (Bruandet et al., 2008; Scarmeas et al., 2006). Thus, education may represent an important additional predictor of neural and behavioral functions underlying aging and neurodegenerative processes. However, education might be associated with potential confounders such as socioeconomic status and life-style, and these factors should be taken into account in future analyses of the TREND study, wherein the average education of 14.3 ± 2.9 years (mean ± SD, n = 325) indicated a high educational level and participation of many academics.

Females showed higher activation within bilateral IFJ, but lower bilateral middle frontal gyri and supramarginal gyri responses compared to males. In part these sex-differences in regional activation might reflect performance differences (females on average slightly outperformed males) related to task performance strategies such as increased switching between word-categories in females compared to males (Lanting

et al., 2009). The neural differences in females compared to males were similar compared to the effects of longer education although females had significantly shorter education (on average 1.3 years less). In this regard, the left inferior frontal gyrus has been shown to be more strongly activated during switching compared to free generation in semantic verbal fluency (Hirshorn and Thompson-Schill, 2006) suggesting that neural differences between males/females, or due to education level, might be related to differences in behavioral strategies. Moreover, the differences in behavioral strategies and underlying neural processing might also partly explain the lack of significant task performance effects on regional activation magnitudes.
Further analysis of the fNIRS data in an extended sample of 654 subjects confirmed the present significant predictors with no major changes in effect sizes (Heinzel et al., 2012). Additionally, increased verbal fluency performance predicted increased left IFJ ($\beta = .09$, $p < .05$) activation in the larger sample consistent with its role for behavioral switching strategies improving performance (Hirshorn and Thompson-Schill, 2006).

For a critical general discussion of the present fNIRS findings a distinction between two possible sources of fNIRS data variability, the previously discussed actual neural and cortical functional hemodynamics on the one hand, and (1) vascular factors and (2) possible task-evoked systemic physiological artifacts on the other hand, might be appropriate.
(1) Functional hemodynamic responses may represent neural correlates of brain functions as neurovascular coupling via astrocytes reliably mediates local increases in cerebral blood volume, flow and oxygenation following neuronal (glutamatergic) synaptic activity (Haydon and Carmignoto, 2006; Iadecola and Nedergaard, 2007; Logothetis, 2002). Thus, compromised vascular function and neurovascular coupling might hinder a straight-forward interpretation of hemodynamic signals as neural activity. For aging as well as hypertension, stroke and AD alterations in hemodynamic responses have been reported (D'Esposito et al., 2003; Girouard and Iadecola, 2006). Vascular risk factors, such as hypertension, diabetes mellitus, cerebrovascular diseases, and hypercholesterolemia increase the AD risk (Barnes and Yaffe, 2011;

Murray et al., 2011). Moreover, cerebral amyloid angiopathy, i.e. the accumulation of amyloid beta-peptides on cerebral blood vessels, is associated with cerebral hypoperfusion and cognitive decline and represents one of the hallmarks of AD (Bell and Zlokovic, 2009). Moreover, smoking, obesity and pharmacological medication may affect vascular function or neurovascular coupling. For instance, aspirin (acetyl-salicylic acid) has antipyretic, anti-inflammatory, analgesic and anti-coagulant effects and is often prescribed as long-term low dose medication to prevent blood clot, strokes and heart attacks (Lewis et al., 1983). Aspirin is an irreversible inhibitor of cyclooxygenase (COX-1, COX-2) involved in the astrocyte-mediated neurovascular coupling which is affected by genetic (Hahn et al., 2011) and pharmacological factors inhibiting COX enzymatic function (Bruhn et al., 2001). Due to regular aspirin intake 13% of the participating subjects were excluded from fNIRS analyses. Since vascular pathologies such as atherosclerosis, coronary heart disease or hypertension have a high incidence in subjects over 50 years of age, future fNIRS analyses will address the issue of altered hemodynamics due to these pathologies or medication using aspirin, antihypertensive drugs and other frequent drugs in the TREND cohort.

(2) Participants of the TREND study may perceive the verbal fluency test as a stressful experimental situation. An example of a verbal instruction during the fNIRS measurements is: "List as many nouns starting with the letter F as you can! (You have 30 seconds.)" First, the task, e.g. generating words on the basis of orthographic criteria, is largely unfamiliar to many participants and unusual in regard to daily cognitive activities. Optimal verbal fluency performance requires efficient organization of verbal retrieval, verbal recall and cognitive self-monitoring, effortful self-initiation, and inhibition of responses as well as other executive functions. While the task may be well suited to identify cognitive deficits (Henry and Crawford, 2004; Henry et al., 2004), its performance may increase stress levels and arousal. Second, subjects (intentionally) participate in a study which might reveal a personal risk or first symptoms of mild cognitive impairment or AD. Accordingly, performing a cognitively demanding task such as the verbal fluency test while brain processes are being monitored may induce additional stress or psychologically challenge subjects, especially when performance is perceived as poor.

Importantly, task-evoked systemic physiological hemodynamic changes, such as changes in heart rate and blood pressure, elicit intra- and extracranial hemodynamic changes in different micro- and macrovascular compartments and, thereby, contaminate fNIRS signals (Kirilina et al., 2012; Minati et al., 2011; Takahashi et al., 2011). Changes in the concentration of oxygenated and deoxygenated hemoglobin, respectively, in the extracortical vascular compartments traversed by near-infrared light affect light intensity at the light detector, confounding functional hemodynamic responses taken to reflect brain activation in cortical gray matter. For the temple region containing the extracranial frontal branches of the temporal artery and vein, fNIRS signal changes have been shown to underlie these extracranial and not cortical hemodynamics (Sato et al., 2011) (see also study #3 below). The (functional) hemodynamic responses and their correlation with age, gender and years of education in the temple region/IFJ should therefore be interpreted with caution.
Verbal fluency task conditions might not only differ in cognitive demands but also in stress levels and arousal, which may contribute to differences in (functional) hemodynamic response magnitudes between task conditions, e.g. "Name as many flowers as you can!" versus "Recite the weekdays!" (semantic fluency condition versus control task).

To conclude, currently fNIRS as used in the present study cannot dissociate between hemodynamic signal changes due to neural processing and variance in this signal due to (aging-related) vascular factors or task-evoked systemic physiological artifacts. For valid interpretations of fNIRS signals on a single subject level and fNIRS as a reliable tool for the personal early prediction of cognitive decline and neurodegeneration, the following issue have to be addressed. The analysis of individual fNIRS data would have to account for (1) vascular (risk) factors affecting (functional) hemodynamic responses, (2) systemic physiological influences on fNIRS signals, (3) individual head morphology and (neuro)anatomy (see study #3 below), and (4) individual (macro-)vascular morphology.
However, vascular risk underlying cerebral hypoperfusion and neural correlates of cognitive processing may in combination represent promising predictors of MCI and

AD. The research on vascular and neural risk of AD will benefit from further fNIRS studies, such as conducted as part of the TREND study, where fNIRS data sets of over 1100 participants have been recorded in only about four months. In the near future, these fNIRS data sets are analyzed by also considering genetic data, blood-based biomarkers, vascular risk profiles, medication, neuropsychological tests, medical histories and other measures, and in regard to longitudinal comparisons of the data of each individual participant.

Study #3

"Variability of (functional) hemodynamics as measured with simultaneous fNIRS and fMRI during intertemporal choice"

The present study first aimed to compare prefrontal fNIRS and fMRI group activation patterns and quantify individual functional hemodynamic responses elicited by intertemporal choice. Second, fNIRS activation magnitudes were investigated for an impact of individual anatomy, i.e. channel-wise scalp-cortex distance and cortical gray matter volume (V_{gray}) simulated to be reached by the near-infrared light. Third, systemic physiological artifacts affecting fNIRS signals were investigated by correlating the fNIRS time series with fMRI signal fluctuations within the scalp. Fourth, prefrontal fNIRS and fMRI activation during intertemporal choice was investigated for consistency in regard to correlations with trait sensitivity to reward (SR), and anatomical and systemic physiological artifacts affecting fNIRS activation-trait associations.

On the group level (20 healthy subjects) the cluster in the right inferior/middle frontal gyrus activation was consistently found by the two simultaneous but independent neuroimaging methods and by previous fNIRS and fMRI studies (McClure et al., 2004; Plichta, 2009). However, fNIRS did not detect additional activation in right inferior frontal gyrus/insula in the temple region as shown by fMRI.

Temporal correlations of the two methods showed wide interindividual and interregional variability. For instance, in the fNIRS peak activation channel (oxy signal, channel #13, middle frontal gyrus) the temporal correlation between the entire time-

series of the fNIRS signal and the corresponding fMRI signal in cortical gray matter simulated to be reached by the near-infrared light, varied from r = -.01 to r = .62 between subjects (mean ± SD; r = .18 ± .17). Additional analyses showed that this interindividual variability was also present for temporal correlations of the event-related average segments of fNIRS and fMRI signals with a mean correlation of r = .57 ± .42 (range: -.76 to .95). A similar degree of interindividual variability in such temporal multimodal correlations has previously been reported for a task battery joint analysis of channel-wise fNIRS-fMRI correlations during motor, response inhibition, visuo-spatial orientation judgment and working memory tasks in right parietal and right prefrontal regions (Cui et al., 2011).

Generally, variance in fNIRS and fMRI activation, respectively, and their multimodal correlations, may be largely determined by interindividual variance in neural task processing and by error variance. For fNIRS, major sources of error variance are given by individual anatomy (Cui et al., 2011; Haeussinger et al., 2011) and by systemic physiological artifacts affecting fNIRS signals and sensitivity for measuring neural activation (Kirilina et al., 2012; Minati et al., 2011; Sato et al., 2011; Takahashi et al., 2011). Individual heads differ in size and shape, but also in the thickness of different layers of skin/muscle tissue (scalp), skull, air (in the frontal sinus region), cerebrospinal fluid, and cortical gray matter. We previously showed that in addition to the scalp-cortex distance (SCD) also the individual composition of these tissue layers varies between subjects and prefrontal/forehead regions (Haeussinger et al., 2011). Moreover, using Monte-Carlo simulations we showed that these anatomical differences are accompanied with differences in the volume of gray matter reached by near-infrared light. The present study showed that these anatomical measures impact the magnitude of fNIRS functional hemodynamic responses, i.e. sensitivity of fNIRS for measuring neural correlates of PFC function.

Thus, on a single subject as well as on the group level activation variance can be partly explained by individual anatomical measures. Complementing previous findings (Cui et al., 2011), SCD and V_{gray} showed the greatest impact in the dorso-medial part of the forehead (superior frontal gyrus) with longest SCD and smallest V_{gray} values.

Here, the fNIRS activation showed correlations of |r| > .05 with these anatomical measures. Therefore, fNIRS studies investigating neural functions in this region, e.g. task-switching in (pre-)supplementary motor cortex (Cutini et al., 2008) should account for the impact of anatomy especially when investigating interindividual or group (male versus females) differences. However, while more pronounced compared to SCD the more specific anatomical measure of V_{gray} showed correlations with fNIRS activation of channels within the inferior/middle frontal gyrus channels where up to 18% of the oxy and up to 41% of the deoxy fNIRS activation variance was explained by V_{gray}. Thus, individual anatomy also plays a role for fNIRS sensitivity in inferior/middle frontal gyrus or (dorso)lateral prefrontal cortex where neural correlates of cognitive functions in healthy subjects, e.g. (Cazzell et al., 2012; Ernst et al., 2011), and psychiatric patients, e.g. (Ehlis et al., 2008; Jourdan Moser et al., 2009), are commonly investigated using fNIRS.

Regarding the impact of (task-evoked) arousal as assessed by fMRI scalp signal fluctuations, pronounced interindividual differences were shown. Positive correlations between fNIRS and scalp fMRI time series were primarily found in the temple region although extending to channels labeled as middle frontal gyrus. In the temple region correlations of up to r = .74 (r = .20 ± .28; mean ± SD between subjects) for the oxy and corresponding negative coefficients for the deoxy fNIRS signal of up to r = -.71 (-.08 ± .30) were found. The fNIRS peak activation channel in the right middle frontal gyrus also showed wide variability between subjects with fMRI scalp signal correlations of up to r = .73 (r = .16 ± .19) for the oxy and r = -.50 (.04 ± .22) for the deoxy fNIRS signal.

While the impact may differ between subjects, systemic physiological artifacts contaminating fNIRS signals are most pronounced within, but not restricted to the temple region. Functional tasks known to elicit changes in heart rate and blood pressure due to (emotional) arousal should be cautiously interpreted, especially when correlating individual factors which might be confounded with these physiological responses (see study #2 and below) or when comparing groups with differences in emotional and/or systemic physiological responses, such as healthy controls versus panic disorder patients or vascular dementia patients. In addition to the implications for future

fNIRS task designs, advances in the fNIRS technology and analysis methods currently address this issue. For instance, fNIRS optodes of multiple distances can enable monitoring and correction of fNIRS data (recorded at 3 cm interoptode distance) using a regressor for hemodynamic signals measured in the scalp (e.g. at 5 mm interoptode distance) (Funane et al., 2012; Gagnon et al., 2012; Saager and Berger, 2008; Takahashi et al., 2011).
Moreover, principal or independent component analyses (PCA, ICA) may be used to decompose a set of different signal components to partly identify and remove signal contribution from extracerebral tissue (Kirkpatrick et al., 1998; Virtanen et al., 2009).

In addition to the present findings of individually weighted and region dependent sources of error variance in fNIRS data, variability differences in neural processing may also underlie differences in cortical functional hemodynamic responses. The present study used an intertemporal choices task as functional paradigm since interindividual differences in neural processing were hypothesized (Peters and Buchel, 2011). Specifically, inferior and middle frontal gyrus have been shown to be involved in cognitive control processes mediating the top-down regulation of the motivational system comprising limbic and other subcortical structures. In this regard the personality construct of trait sensitivity to reward was hypothesized to be associated with a decrease in the activation in inferior/middle frontal gyrus underlying cognitive control. Using fMRI we confirmed this hypothesis as during the evaluation and choice of reward options also involving immediate rewards the activation in this cortical region showed a negative correlation with the trait. When both reward options of choice were only available after a delay of a minimum two weeks the correlation was not significant. However, using fNIRS no association between the trait and cortical activation was detected. Thus, the previously identified individually weighted sources of error variance of fNIRS data present in inferior/middle frontal gyrus might have affected or biased the trait-activation association which exhibited a non-significant positive correlation. While the detection of this association might not have been robust enough against anatomical error variance (V_{gray}) or the distance to the correlation voxel cluster identified with fMRI, systemic physiological artifacts might

have biased the negative correlation shown with fMRI in the positive direction. Some evidence for such a confounding association between trait sensitivity to reward with systemic physiological effects during the reward-based decision making task was revealed by positive correlations between the trait and the time series correlations between fNIRS and scalp fMRI signal fluctuations. Thus, in some subjects with high trait SR scores extracerebral hemodynamics might have produced an increase in fNIRS signals although, as suggested by the fMRI findings, the functional hemodynamic response due to neural processing would be relatively decreased compared to subjects with low trait SR and putatively increased cognitive control functions during intertemporal choice.

The present study focused on right prefrontal activation during intertemporal choice and an impact of trait SR on the processing of subcortical structures or cortico-limbic interactions was therefore beyond the scope of the present study. However, the missing link between trait SR and behavioral consequences in intertemporal choice behavior or decision times might be explained by further analyses of the ventral striatum, posterior cingulate cortex and medial prefrontal cortex, which have been postulated as a valuation network involved in intertemporal choice (Kable and Glimcher, 2007). Thus, behavioral consequences of the trait SR modulation of cognitive control via right inferior/middle frontal gyrus engagement might emerge from the interaction of this prefrontal region with the subcortical valuation network. Thereby, the personality construct of trait SR could be implemented into distributed and interacting neural networks, which underlie reward valuation, prospection and decision-making involved in intertemporal choice (Peters and Buchel, 2011).

Concluding remarks

As concluding remarks of this cumulative dissertation I would like to point out the highly innovative methods, interdisciplinary approaches and the increasingly growing knowledge in current neuroscience. While largely unforeseeable in its specific nature, major advances and discoveries in the next decade(s) can be expected, which may partly be due to a major paradigm shift in neuroscience: The immense computational and methodological advances and the massive increase in access to genetic, cellular, biomarker, structural and functional neuroimaging, behavioral, demographic and epidemiological data in animal models and humans enable research and innovative ideas which may be quantified by the publication of over 60,000 neuroscientific papers per year (Waldrop, 2012).

Future developments in information technologies might help to link new findings to current knowledge, generate new testable hypotheses, and integrate new findings into a (simulated) working model of the human brain and brain-related diseases (Markram, 2012).

References (General discussion)

Apud, J.A., Mattay, V., Chen, J., Kolachana, B.S., Callicott, J.H., Rasetti, R., Alce, G., Iudicello, J.E., Akbar, N., Egan, M.F., Goldberg, T.E., Weinberger, D.R., 2007. Tolcapone improves cognition and cortical information processing in normal human subjects. Neuropsychopharmacology 32, 1011-1020.

Aron, A.R., Poldrack, R.A., 2006. Cortical and subcortical contributions to Stop signal response inhibition: role of the subthalamic nucleus. The Journal of neuroscience : the official journal of the Society for Neuroscience 26, 2424-2433.

Barch, D.M., Ceaser, A., 2012. Cognition in schizophrenia: core psychological and neural mechanisms. Trends Cogn Sci 16, 27-34.

Barnes, D.E., Yaffe, K., 2011. The projected effect of risk factor reduction on Alzheimer's disease prevalence. Lancet Neurol 10, 819-828.

Bell, R.D., Zlokovic, B.V., 2009. Neurovascular mechanisms and blood-brain barrier disorder in Alzheimer's disease. Acta Neuropathol 118, 103-113.

Bertolino, A., Caforio, G., Blasi, G., De Candia, M., Latorre, V., Petruzzella, V., Altamura, M., Nappi, G., Papa, S., Callicott, J.H., Mattay, V.S., Bellomo, A., Scarabino, T., Weinberger, D.R., Nardini, M., 2004. Interaction of COMT

(Val(108/158)Met) genotype and olanzapine treatment on prefrontal cortical function in patients with schizophrenia. Am J Psychiatry 161, 1798-1805.
Bonifacio, M.J., Palma, P.N., Almeida, L., Soares-da-Silva, P., 2007. Catechol-O-methyltransferase and its inhibitors in Parkinson's disease. CNS Drug Rev 13, 352-379.
Bruandet, A., Richard, F., Bombois, S., Maurage, C.A., Masse, I., Amouyel, P., Pasquier, F., 2008. Cognitive decline and survival in Alzheimer's disease according to education level. Dement Geriatr Cogn Disord 25, 74-80.
Bruhn, H., Fransson, P., Frahm, J., 2001. Modulation of cerebral blood oxygenation by indomethacin: MRI at rest and functional brain activation. J Magn Reson Imaging 13, 325-334.
Bush, G., Shin, L.M., 2006. The Multi-Source Interference Task: an fMRI task that reliably activates the cingulo-frontal-parietal cognitive/attention network. Nat Protoc 1, 308-313.
Caamano-Isorna, F., Corral, M., Montes-Martinez, A., Takkouche, B., 2006. Education and dementia: a meta-analytic study. Neuroepidemiology 26, 226-232.
Cazzell, M., Li, L., Lin, Z.J., Patel, S.J., Liu, H., 2012. Comparison of neural correlates of risk decision making between genders: An exploratory fNIRS study of the Balloon Analogue Risk Task (BART). Neuroimage 62, 1896-1911.
Cools, R., D'Esposito, M., 2011. Inverted-U-shaped dopamine actions on human working memory and cognitive control. Biological psychiatry 69, e113-125.
Cui, X., Bray, S., Bryant, D.M., Glover, G.H., Reiss, A.L., 2011. A quantitative comparison of NIRS and fMRI across multiple cognitive tasks. Neuroimage 54, 2808-2821.
Cutini, S., Scatturin, P., Menon, E., Bisiacchi, P.S., Gamberini, L., Zorzi, M., Dell'Acqua, R., 2008. Selective activation of the superior frontal gyrus in task-switching: an event-related fNIRS study. Neuroimage 42, 945-955.
D'Esposito, M., Deouell, L.Y., Gazzaley, A., 2003. Alterations in the BOLD fMRI signal with ageing and disease: a challenge for neuroimaging. Nat Rev Neurosci 4, 863-872.
Durstewitz, D., Seamans, J.K., 2008. The dual-state theory of prefrontal cortex dopamine function with relevance to catechol-o-methyltransferase genotypes and schizophrenia. Biological psychiatry 64, 739-749.
Ehlis, A.C., Bahne, C.G., Jacob, C.P., Herrmann, M.J., Fallgatter, A.J., 2008. Reduced lateral prefrontal activation in adult patients with attention-deficit/hyperactivity disorder (ADHD) during a working memory task: a functional near-infrared spectroscopy (fNIRS) study. J Psychiatr Res 42, 1060-1067.
Ernst, L.H., Plichta, M.M., Lutz, E., Zesewitz, A.K., Tupak, S.V., Dresler, T., Ehlis, A.C., Fallgatter, A.J., 2011. Prefrontal activation patterns of automatic and regulated approach-avoidance reactions - A functional near-infrared spectroscopy (fNIRS) study. Cortex.
Fallgatter, A.J., Bartsch, A.J., Herrmann, M.J., 2002. Electrophysiological measurements of anterior cingulate function. J Neural Transm 109, 977-988.
Farrell, S.M., Tunbridge, E.M., Braeutigam, S., Harrison, P.J., 2012. COMT Val(158)Met genotype determines the direction of cognitive effects produced by catechol-O-methyltransferase inhibition. Biological psychiatry 71, 538-544.

Funane, T., Atsumori, H., Kiguchi, M., Tanikawa, Y., Okada, E., 2012. Dynamic phantom with two stage-driven absorbers for mimicking hemoglobin changes in superficial and deep tissues. J Biomed Opt 17, 047001.

Gagnon, L., Cooper, R.J., Yucel, M.A., Perdue, K.L., Greve, D.N., Boas, D.A., 2012. Short separation channel location impacts the performance of short channel regression in NIRS. Neuroimage 59, 2518-2528.

Girouard, H., Iadecola, C., 2006. Neurovascular coupling in the normal brain and in hypertension, stroke, and Alzheimer disease. J Appl Physiol 100, 328-335.

Haeussinger, F.B., Heinzel, S., Hahn, T., Schecklmann, M., Ehlis, A.C., Fallgatter, A.J., 2011. Simulation of near-infrared light absorption considering individual head and prefrontal cortex anatomy: implications for optical neuroimaging. PLoS One 6, e26377.

Hahn, T., Heinzel, S., Plichta, M.M., Reif, A., Lesch, K.P., Fallgatter, A.J., 2011. Neurovascular coupling in the human visual cortex is modulated by cyclooxygenase-1 (COX-1) gene variant. Cereb Cortex 21, 1659-1666.

Hall, C.B., Derby, C., LeValley, A., Katz, M.J., Verghese, J., Lipton, R.B., 2007. Education delays accelerated decline on a memory test in persons who develop dementia. Neurology 69, 1657-1664.

Haydon, P.G., Carmignoto, G., 2006. Astrocyte control of synaptic transmission and neurovascular coupling. Physiol Rev 86, 1009-1031.

Heinzel, S., Metzger, F., Ehlis, A.-C., Korell, R., Alboji, A., Haeussinger, F.B., Hagen, K., Eschweiler, G.W., Berg, D., Fallgatter, A.J., 2012. The impact of age and gender on prefrontal cortex processing during verbal fluency: an fNIRS study. Human Brain Mapping Conference, Beijing, China.

Henry, J.D., Crawford, J.R., 2004. A meta-analytic review of verbal fluency performance following focal cortical lesions. Neuropsychology 18, 284-295.

Henry, J.D., Crawford, J.R., Phillips, L.H., 2004. Verbal fluency performance in dementia of the Alzheimer's type: a meta-analysis. Neuropsychologia 42, 1212-1222.

Hipp, G., Pieri, V., Vaillant, M., Diederich, N., 2009. Verbal fluency and psychomotor speed in early Parkinson's disease: is there a link? Parkinsonism & Related Disorders 15.

Hirshorn, E.A., Thompson-Schill, S.L., 2006. Role of the left inferior frontal gyrus in covert word retrieval: neural correlates of switching during verbal fluency. Neuropsychologia 44, 2547-2557.

Iadecola, C., Nedergaard, M., 2007. Glial regulation of the cerebral microvasculature. Nat Neurosci 10, 1369-1376.

Jack, C.R., Jr., Knopman, D.S., Jagust, W.J., Shaw, L.M., Aisen, P.S., Weiner, M.W., Petersen, R.C., Trojanowski, J.Q., 2010. Hypothetical model of dynamic biomarkers of the Alzheimer's pathological cascade. Lancet Neurol 9, 119-128.

Jourdan Moser, S., Cutini, S., Weber, P., Schroeter, M.L., 2009. Right prefrontal brain activation due to Stroop interference is altered in attention-deficit hyperactivity disorder - A functional near-infrared spectroscopy study. Psychiatry Res 173, 190-195.

Kirilina, E., Jelzow, A., Heine, A., Niessing, M., Wabnitz, H., Bruhl, R., Ittermann, B., Jacobs, A.M., Tachtsidis, I., 2012. The physiological origin of task-evoked

systemic artefacts in functional near infrared spectroscopy. Neuroimage 61, 70-81.
Kirkpatrick, P.J., Smielewski, P., Al-Rawi, P., Czosnyka, M., 1998. Resolving extra- and intracranial signal changes during adult near infrared spectroscopy. Neurol Res 20 Suppl 1, S19-22.
Klumpp, H., Deldin, P., 2010. Review of brain functioning in depression for semantic processing and verbal fluency. Int J Psychophysiol 75, 77-85.
Ko, J.H., Antonelli, F., Monchi, O., Ray, N., Rusjan, P., Houle, S., Lang, A.E., Christopher, L., Strafella, A.P., 2012. Prefrontal dopaminergic receptor abnormalities and executive functions in Parkinson's disease. Hum Brain Mapp.
Koob, G.F., Volkow, N.D., 2010. Neurocircuitry of addiction. Neuropsychopharmacology 35, 217-238.
Lanting, S., Haugrud, N., Crossley, M., 2009. The effect of age and sex on clustering and switching during speeded verbal fluency tasks. J Int Neuropsychol Soc 15, 196-204.
Lewis, H.D., Jr., Davis, J.W., Archibald, D.G., Steinke, W.E., Smitherman, T.C., Doherty, J.E., 3rd, Schnaper, H.W., LeWinter, M.M., Linares, E., Pouget, J.M., Sabharwal, S.C., Chesler, E., DeMots, H., 1983. Protective effects of aspirin against acute myocardial infarction and death in men with unstable angina. Results of a Veterans Administration Cooperative Study. N Engl J Med 309, 396-403.
Logothetis, N.K., 2002. The neural basis of the blood-oxygen-level-dependent functional magnetic resonance imaging signal. Philos Trans R Soc Lond B Biol Sci 357, 1003-1037.
Markram, H., 2012. The Human Brain Project. Scientific American 306, 50-55.
McClure, S.M., Laibson, D.I., Loewenstein, G., Cohen, J.D., 2004. Separate neural systems value immediate and delayed monetary rewards. Science 306, 503-507.
Minati, L., Kress, I.U., Visani, E., Medford, N., Critchley, H.D., 2011. Intra- and extra-cranial effects of transient blood pressure changes on brain near-infrared spectroscopy (NIRS) measurements. J Neurosci Methods 197, 283-288.
Murray, I.V., Proza, J.F., Sohrabji, F., Lawler, J.M., 2011. Vascular and metabolic dysfunction in Alzheimer's disease: a review. Exp Biol Med (Maywood) 236, 772-782.
Niendam, T.A., Laird, A.R., Ray, K.L., Dean, Y.M., Glahn, D.C., Carter, C.S., 2012. Meta-analytic evidence for a superordinate cognitive control network subserving diverse executive functions. Cogn Affect Behav Neurosci.
Ownby, R.L., Crocco, E., Acevedo, A., John, V., Loewenstein, D., 2006. Depression and risk for Alzheimer disease: systematic review, meta-analysis, and metaregression analysis. Arch Gen Psychiatry 63, 530-538.
Peters, J., Buchel, C., 2011. The neural mechanisms of inter-temporal decision-making: understanding variability. Trends Cogn Sci 15, 227-239.
Plichta, M.M., 2009. Neural correlates of delay discounting: Effects of dopamine bioavailability and implications for attention-deficit/hyperactivity disorder (ADHD). Doctoral thesis. University of Wuerzburg.

Rusz, J., Cmejla, R., Ruzickova, H., Ruzicka, E., 2011. Quantitative acoustic measurements for characterization of speech and voice disorders in early untreated Parkinson's disease. J Acoust Soc Am 129, 350-367.

Saager, R., Berger, A., 2008. Measurement of layer-like hemodynamic trends in scalp and cortex: implications for physiological baseline suppression in functional near-infrared spectroscopy. J Biomed Opt 13, 034017.

Sato, H., Obata, A.N., Moda, I., Ozaki, K., Yasuhara, T., Yamamoto, Y., Kiguchi, M., Maki, A., Kubota, K., Koizumi, H., 2011. Application of near-infrared spectroscopy to measurement of hemodynamic signals accompanying stimulated saliva secretion. J Biomed Opt 16, 047002.

Scarmeas, N., Albert, S.M., Manly, J.J., Stern, Y., 2006. Education and rates of cognitive decline in incident Alzheimer's disease. J Neurol Neurosurg Psychiatry 77, 308-316.

Schroeter, M.L., Vogt, B., Frisch, S., Becker, G., Barthel, H., Mueller, K., Villringer, A., Sabri, O., 2012. Executive deficits are related to the inferior frontal junction in early dementia. Brain 135, 201-215.

Sharp, E.S., Gatz, M., 2011. Relationship Between Education and Dementia An Updated Systematic Review. Alzheimer Disease & Associated Disorders 25, 289-304.

Stein, J.L., Hua, X., Lee, S., Ho, A.J., Leow, A.D., Toga, A.W., Saykin, A.J., Shen, L., Foroud, T., Pankratz, N., Huentelman, M.J., Craig, D.W., Gerber, J.D., Allen, A.N., Corneveaux, J.J., Dechairo, B.M., Potkin, S.G., Weiner, M.W., Thompson, P., 2010. Voxelwise genome-wide association study (vGWAS). Neuroimage 53, 1160-1174.

Stern, Y., 2006. Cognitive reserve and Alzheimer disease. Alzheimer Dis Assoc Disord 20, S69-74.

Takahashi, T., Takikawa, Y., Kawagoe, R., Shibuya, S., Iwano, T., Kitazawa, S., 2011. Influence of skin blood flow on near-infrared spectroscopy signals measured on the forehead during a verbal fluency task. Neuroimage 57, 991-1002.

Troyer, A.K., Moscovitch, M., Winocur, G., 1997. Clustering and switching as two components of verbal fluency: Evidence from younger and older healthy adults. Neuropsychology 11, 138-146.

Tucker, A.M., Stern, Y., 2011. Cognitive reserve in aging. Curr Alzheimer Res 8, 354-360.

Virtanen, J., Noponen, T., Merilainen, P., 2009. Comparison of principal and independent component analysis in removing extracerebral interference from near-infrared spectroscopy signals. J Biomed Opt 14, 054032.

Waldrop, M.M., 2012. Computer modelling: Brain in a box. Nature 482, 456-458.

Woodward, N.D., Jayathilake, K., Meltzer, H.Y., 2007. COMT val108/158met genotype, cognitive function, and cognitive improvement with clozapine in schizophrenia. Schizophr Res 90, 86-96.

Acknowledgements

The studies of the past years and their interdisciplinary nature would not have been possible without the practical, intellectual and motivational support of many individual persons whom I would like to thank.

First of all I would like to thank Prof. Dr. Andreas J. Fallgatter for the supervision of this thesis and his encouraging support, also in regard to several other interdisciplinary neuroimaging and clinical studies and ongoing scientific projects. Thank you for introducing me into the field of Imaging Genetics and clinical research.

I appreciate the supervision of Prof. Dr. Dr. h.c. Martin Heisenberg facilitating this external cumulative dissertation at the Faculty of Biology of the University of Würzburg. Moreover, I like to thank for his inspiration during my study of biology linking molecular neurogenetics to neurobehavioral phenotypes and to seek out from biology to other disciplines such as psychology and medicine.

I am thankful for the contributions of all co-authors supporting or enabling the studies included in this cumulative dissertation.

Especially, I am grateful for the reliable neuroscientific and methodological EEG and fNIRS expertise of Dr. Ann-Christine Ehlis, her feedback on this thesis and her organizational and motivational skills.

I thank my colleagues in the lab of Psychophysiology and Optical Imaging at the Department of Psychiatry and Psychotherapy in Tübingen for the friendly (working) atmosphere, and in particular Dipl.-Phys. Florian Häußinger for fruitful discussions on the physical basis of neuroimaging methods as well as his programming talents.

I thank Dr. Tim Hahn for the many inspiring, exciting and competent discussions about neuroscience, psychology and statistics, and the shared intrinsic motivation and enthusiasm which stimulated many of our projects.

Moreover, I thank Dr. Tom Baden for the many interesting discussions about biology and its many aspects in science and life, and his feedback on this thesis.

I wish to express my special gratitude to my wonderful parents and my sister, who always supported me in my life and my work.

List of scientific publications

Publications as First Author *equal contribution; Impact factor (IF)

1. **Heinzel, S.***, Haeussinger, F.B.*, Hahn, T., Ehlis, A.C., Plichta, M.M., Fallgatter, A.J., 2013. Variability of (functional) hemodynamics as measured with simultaneous fNIRS and fMRI during intertemporal choice. *Neuroimage* 2013. Jan 8;71C:125-134. doi: 10.1016/j.neuroimage.2012.12.074. [Epub ahead of print]. **IF: 5.9**

2. **Heinzel, S.**, Dresler, T., Baehne, C.G., Heine, M., Boreatti-Hummer, A., Jacob, C.P., Renner, T.J., Reif, A., Lesch, K.P., Fallgatter, A.J.*, Ehlis, A.C.*, 2012. COMT x DRD4 Epistasis Impacts Prefrontal Cortex Function Underlying Response Control. *Cerebral Cortex*, 2012 May 28 [Epub ahead of print]. **IF: 6.5**

3. **Heinzel, S.**, Metzger, F.G., Ehlis, A.C., Korell, R., Alboji, A., Haeussinger, F.B., Hagen, K., Maetzler, W., Eschweiler, G.W., Berg, D., Fallgatter, A.J., 2012. Aging-related cortical reorganization of verbal fluency processing: a functional near-infrared spectroscopy study. *Neurobiology of Aging*, Feb;34(2):439-50. doi: 10.1016/j.neurobiolaging.2012.05.021. Epub 2012 Jul 6. **IF: 6.2**

4. Hahn, T.*, **Heinzel, S.***, Plichta, M.M., Reif, A., Lesch, K.P., Fallgatter, A.J., 2011. Neurovascular coupling in the human visual cortex is modulated by cyclooxygenase-1 (COX-1) gene variant. *Cerebral Cortex* 21, 1659-1666. **IF: 6.5**

5. Haeussinger, F.B.*, **Heinzel, S.***, Hahn, T., Schecklmann, M., Ehlis, A.C., Fallgatter, A.J., 2011. Simulation of near-infrared light absorption considering individual head and prefrontal cortex anatomy: implications for optical neuroimaging. *PLoS One* 6, e26377. **IF: 4.4**

6. Hahn, T.*, **Heinzel, S.***, Dresler, T.*, Plichta, M.M., Renner, T.J., Markulin, F., Jakob, P.M., Lesch, K.P., Fallgatter, A.J., 2010. Association between reward-related activation in the ventral striatum and trait reward sensitivity is moderated by dopamine transporter genotype. *Human Brain Mapping* 32, 1557-1565. **IF: 5.9**

Publications as Co-author

7. Lesch, K.P., Selch, S., Renner, T.J., Jacob, C., Nguyen, T.T., Hahn, T., Romanos, M., Walitza, S., Shoichet, S., Dempfle, A., Heine, M., Boreatti-Hummer, A., Romanos, J., Gross-Lesch, S., Zerlaut, H., Wultsch, T., **Heinzel, S.**, Fassnacht, M., Fallgatter, A., Allolio, B., Schafer, H., Warnke, A., Reif, A., Ropers, H.H., Ullmann, R., 2011. Genome-wide copy number variation analysis in attention-deficit/hyperactivity disorder: association with neuropeptide Y gene dosage in an extended pedigree. *Molecular Psychiatry* 16, 491-503. **IF: 13.7**

8. Dresler, T., Ehlis, A.C., **Heinzel, S.**, Renner, T.J., Reif, A., Baehne, C.G., Heine, M., Boreatti-Hummer, A., Jacob, C.P., Lesch, K.P., Fallgatter, A.J., 2010. Dopamine transporter (SLC6A3) genotype impacts neurophysiological correlates of cognitive response control in an adult sample of patients with ADHD. *Neuropsychopharmacology* 35, 2193-2202. **IF: 8.0**

9. Hahn, T., Dresler, T., Ehlis, A.C., Plichta, M.M., **Heinzel, S.**, Polak, T., Lesch, K.P., Breuer, F., Jakob, P.M., Fallgatter, A.J., 2009. Neural response to reward anticipation is modulated by Gray's impulsivity. Neuroimage 46, 1148-1153. **IF: 5.9**

10. Plichta, M.M., **Heinzel, S.**, Ehlis, A.C., Pauli, P., Fallgatter, A.J., 2007. Model-based analysis of rapid event-related functional near-infrared spectroscopy (NIRS) data: a parametric validation study. *Neuroimage* 35, 625-634. **IF: 5.9**

Submitted

11. Hahn, T., **Heinzel, S.**, Notebaert, K., Dresler, T., Reif, A., Lesch, K.P., Jakob, P.M., Windmann, S., Fallgatter, A.J.: The tricks of the trait: Neural implementation of personality varies with genotype-dependent efficiency of serotonergic neurotransmission; *in Revision*.

12. Plichta, M.M.*****, **Heinzel, S.***, Gerdes, A.B.M., Ehlis, A.C., Schecklmann, M., Reif, A., Erdfelder, E., Dan, I., Tsuzuki, D., Lesch, K.P., Grön, G., Pauli, P., Fallgatter, A.J.: Impulsivity-related prefrontal activation during Reward Discounting is moderated by *COMT* Val^{158}Met polymorphism; *submitted*.

List of major conference contributions

- Invited talk at the "Psychologie & Gehirn" Conference 2012 (Jena, Germany)
- Invited talk at the "Human Brain Mapping" Conference 2010 (Barcelona, Spain)
- Human Brain Mapping Conference 2012 (Beijing, China), 2010 (Barcelona, Spain), 2009 (San Francisco, USA); Poster
- Forum of European Neuroscience (FENS) 2010 (Amsterdam, Netherlands); Poster
- International Congress of Psychology 2008 (Berlin, Germany); Poster

Curriculum vitae (CV)

Sebastian Heinzel, Feb. 2013

Education

Doctoral thesis (Dr. rer. nat., *Magna cum Laude*)
03/2008 – 01/2013 Multimodal neuroimaging of prefrontal cortex (dys)function: EEG, fNIRS, fNIRS-fMRI and Imaging Genetics approaches
Supervisors:
Prof. Dr. Andreas J. Fallgatter
Dept. of Psychiatry & Psychotherapy, University of Tübingen, Germany
Prof. Dr. Dr. h.c. Martin Heisenberg
Rudolf Virchow Center, University of Würzburg, Germany

Diploma (biology)
04/2007 – 02/2008 Thesis: Neural correlates of delay discounting and genetic polymorphisms affecting dopaminergic transmission. (grade: 1.3, supervisors: A.J. Fallgatter, M. Heisenberg)

10/2002 – 02/2008 Biology (Diploma: Neurobiology (grade: 1.3), Genetics (1.7), Biological Psychology (1.0)): University of Würzburg

10/2001 – 9/2002 Physics (Diploma): University of Bonn, Germany

06/2000 Abitur, Otto-Kühne-Schule, Bonn, Germany

Research assistant

02/2013 – Postdoc, Dept. of Neurodegeneration, Hertie Institute for Clinical Brain Research (HIH), Tübingen, Germany
Lab: Prof. Daniela Berg

10/2010 – 01/2013 German Center for Neurodegenerative diseases (DZNE), Tübingen, and Dept. of Psychiatry and Psychotherapy, University of Tübingen

09/2010 – 10/2010 Dept. of Psychiatry and Psychotherapy, University of Tübingen

04/2008 – 08/2010 Dept. of Psychiatry, Psychosomatics and Psychotherapy, University of Würzburg

Scholarships

09/2009 – 09/2010 Scholarship of the Federal Ministry of Economy and Technology (BMWi)

Grant writing

Writing of the grant proposal for the "EXIST" support program of the Federal Ministry of Economy and Technology (BMWi): University-based business start-up
Submitted by Prof. A.J. Fallgatter (78.400€ approved; 2009-2010)

Teaching skills

06/2012 – Speaker/Member of the Organizational Board of the TREND-study (**T**übinger evaluation of **R**isk factors for **E**arly detection of **N**euro**D**egeneration)

08/2010 – Associate editor for the journal "Frontiers in Human Neuro-science"; Host of the Special Topic "Triadic interactions of brain activation, (behavioral) trait and genotype"
In collaboration with Dr. Tim Hahn (University of Frankfurt) and Dr. Michael M. Plichta (Central Institute of Mental Health, Mannheim)

01/2009 – 10/2010 Practical and analysis supervision of diploma theses:

- Florian Häußinger (Physicist):
  Quantitative comparison of methods measuring hemo-dynamic responses – a simultaneous fMRI and fNIRS-study.

- Bastian Schiller (Psychologist):
  Neural correlates and modulation of decision-making under risk – a neuro-navigated TMS and fNIRS-study.

06/2009 – Practical and analysis supervision of medical doctoral theses

- Heiner Gieseke (Cand. med.): fNIRS, ADHD, *COMT*-genotype, delay discounting

- Robert Korell (Cand. med.): TREND-study, fNIRS, demographics

- Ahmed Alboji (Cand. med.): TREND-study, fNIRS, pharmacology